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Postharvest investigations into chlorophyll fluorescence and low temperature injury in cut roses (*Rosa hybrida* L.)

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Στη μνήμη της μάνας μου Ειρήνης

The roots of education are bitter but the fruit is sweet

Aristotle

ABSTRACT

This is one of the first studies on the relationship between pre-harvest environmental conditions found in the Mediterranean and postharvest characteristics of cut roses (*Rosa hybrida* L.). Effects of storage temperature on vase life parameters were also studied for roses grown all year round. The postharvest storage of roses at low temperature is a useful practice, in terms of market flow regulation. However, a reduction in vase life and loss of flower quality has been recorded after storage due to Low Temperature Injury (LTI). LTI of roses is difficult to assess by visual observation. Relative chlorophyll fluorescence (F_v/F_m), which is a non-invasive technique that provides an index of stress effects on photosystem II (PS II) activity, was used to investigate LTI in roses. The plant growth regulator abscisic acid (ABA) can cause physiological responses that protect plants against CI or LTI. The overall objectives of this study were firstly to evaluate the pre-harvest environmental conditions affecting vase life and secondly to evaluate novel potential ABA treatments to protect cut roses against LTI.

Vase life durations and F_v/F_m ratios measured after low temperature storage for 'First Red' and 'Akito' roses were seasonally dependant. Vase lives of roses grown during winter were significantly ($P \leq 0.001$) shorter compared to roses grown during the rest of the year. In autumn and winter experiments F_v/F_m ratios were generally reduced following storage at 1°C, suggesting LTI of roses. Thus, the fall of F_v/F_m was due to an interaction of growing season and storage at 1°C. However, in second year experiments, growing temperature and PFD were relatively higher and, as a result, F_v/F_m did not decline for 'Akito' roses after low temperature storage, indicating a strong influence of environmental conditions. Higher PFD and temperature glasshouse during the year were positively and significantly correlated with maintenance of post-storage F_v/F_m ratios and longer vase life. It is suggested that shorter vase lives and lower post-storage F_v/F_m values after storage at 1°C are consequences of reduced photosynthesis and smaller carbohydrate pools in winter-harvested roses. Because of the lack of correlation between F_v/F_m and post-storage vase life, it is concluded that the CF parameter F_v/F_m is not a practical index for assessing LTI in cold-stored roses. Growing roses in autumn and winter months

increased flower blueing in red petals of 'First Red' roses and prevented flower opening for both cultivars.

ABA applied as pulse treatment before storage or as vase solution during vase life generally improved vase life parameters. Pulsing 'Akito' roses with 10^{-1} M ABA before storage increased vase life and inhibited bent neck incidence. Also, the presence of ABA in vase solution increased vase life after storage at 1°C, reducing vase solution usage during vase life. Similarly, the synthetic ABA analogue PBI-365, as vase solution ingredient, was effective in extending vase life and reducing transpiration rates in roses after low temperature storage. Increased ABA levels were detected in leaves and petals using HPLC when roses were treated with exogenous ABA before storage and during vase life. Thus, it was assumed that ABA or PBI-365 acted on guard cells by causing stomatal closure. Electrolyte leakage and lipid peroxidation, measured after storage at 1°C, were markedly reduced by application of ABA. Both pulse ABA treatment and vase solutions containing ABA helped to recover F_v/F_m during vase life. Moreover, addition of PBI-365 in vase solution reduced the degree of lipid peroxidation in leaves and petals after storage at 1°C. These observations indicated a protection role of ABA against LTI for roses, which has also been observed in other crops.

Further research at the cellular and/or molecular level may help in better understanding the physiological responses of roses to seasonal variation during the year and LTI. In addition, work is also required to look at the ABA and PBI-365 mode of action in roses. Additional research using a wide range of ABA concentrations and assays using exogenous radio-labelled ABA may help to better understand the nature of ABA efficacy.

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LIST OF ABBREVIATIONS

ABA	abscisic acid
ACC	aminocyclopropane-1-carboxylic acid
AI	active ingredient
ANOVA	analysis of variance
ATP	adenosine triphosphate
BHT	2,6-di-t-butyl-4-methyl-phenol
ca.	approximately
CAT	catalase
CF	chlorophyll fluorescence
cfu	colony forming units
CHS	chalcone synthase
CI	chilling injury
cm	centimetre
CRD	completely randomised design
cv.	cultivar
°C	degrees Celsius
d	day
DICA	dichlorocyanuric acid
DW	dry weight
EC	electrical conductivity
e.g.	for example
EL	electrolyte leakage
et al.	and others
F.A.A	formalin – acetic acetic – ethanol
F_v	variable fluorescence
F_m	maximum fluorescence
F_0	minimum fluorescence
F_v/F_m	relative fluorescence
FW	fresh weight
g	gram
GH	glasshouse

GLM	general linear model
h	hour
HPLC	high performance liquid chromatography
IAA	indole-3-acetic acid
i.e.	that is
ivr	invertase
l	litre
LOX	lipoxygenase
LP	low pressure
LPS	lateral phase seperation
LSD	least significant difference
LTI	low temperature injury
m	metre
M	molar (moles/L)
MDA	malondialdehyde
1-MCP	methylcyclopropene
µg	microgram (10^{-6} kg)
µl	microlitre (10^{-6} L)
µM	micromole (10^{-6} mol)
mg	milligram (10^{-3} g)
min	minutes
ml	millilitre (10^{-3} l)
mm	millimetre (10^{-3} m)
mM	millimolar (10^{-3} M)
mol	mole
mS	milli-siemens
n	number of observations comprising a value
NCER	plant net CO_2 exchange rate
nl	nanolitre (10^{-9} L)
nm	nanometre
<	less than
n.s.	not significant at $P \leq 0.05$
O.D.	optical density

P	probability
PAL	phenylalanine ammonia lyase
PBI	plant biotechnology institute
pers. comm.	personal communication
PFD	photon flux density
pH	relative proton concentration in a solution
PPFD	photosynthetic photon flux density
PSII	photosystem 2
%	percent
±	plus or minus
®	registered trademark
R.F.W.	relative fresh weight
RH	relative humidity
Rpm	revolutions per minute
s	second
SAM	S-adenosylmethionine
s.e.	standard error
sp. /spp.	species
STS	silver thiosulfate virus
UV-C	short-wave ultraviolet
vs.	versus
viz.	that is
w	weight

CHAPTER 1

GENERAL INTRODUCTION

1.1 BACKGROUND

Cut flowers have played an important role in human society for thousands of years. For a range of purposes, consumers have used cut flowers; for their medicinal value, and as food, but most importantly as a welcoming and beautiful gift by which people communicate with one another. The first cultivated roses appeared in Asian gardens more than 5,000 years ago. Roses were introduced into Europe during the Roman Empire, where they were used for ornamental purposes. In Crete, the Minoans of the middle Minoan Knossos period were aware of roses as an example was found on a fresco from 1600 BC. The rose had six petals, and, thus, could have been the rose R. X. Richardie, which was the rose of Ethiopian churches long known as *R. Sancta* (thought to be a hybrid of *R. gallica*.)

Today, flowers are grown for commercial purposes in many countries. The Netherlands plays the pivotal role in the world cut flower trade. Holland is the largest producer of cut flowers in the world and is the world's largest flower exporter through specialised flower auctions, such as that at Aalsmeer. Cut flowers are exported from Holland to most European countries, the United States of America and Russia (Appendix 1, Table A1.1). Roses are of the most important flower exports of Holland followed by tulip, chrysanthemum and gerberas (Appendix 1, Table A1.2). In Crete, a significant amount of cut flowers were produced from 1984 until 2000 (Table 1.1). Some of these flowers were exported from Crete to Europe and Japan from 1980 to 1985. Thereafter, exports were halted due to high transport costs and problems with vase life. Over the last years cut flower exports have increased but their importance is still low (Table 1.2).

The postharvest life of cut flowers (the vase life) is deemed as starting from the time at which a flower is placed within a vase. Vase life ends when advance signs of deterioration are visible on flower and/or leaves (Mayak and Halevy, 1974) but this is highly subjective. Vase life is a characteristic feature of each species and cultivar and is strongly dependent on cultural conditions and handling after harvest. The vase

life of cut flowers often depends on the season. Pre-harvest environmental conditions, such as temperature, relative humidity, light intensity and photoperiod during cultivation, significantly influence the vase life of cut roses (Shin *et al.*, 2001).

Like other cut flowers, roses, are highly perishable. As cut roses move through the postharvest chain, from grower to consumer, floral organs continue to actively metabolise. As with other cut flower species, suitable postharvest storage and transport procedures are required for maintenance of rose quality during transport from the production site (e.g. Crete) to the marketplace (Leonard *et al.*, 2001). Cut roses are stored at low temperature to reduce postharvest respiration and transpiration, and to prolong longevity. Holding cut roses at low temperature seems attractive, as storage offers greater flexibility in terms of selling time. The most evident effect of cold storage is a decrease in metabolic activity. However, low temperature storage (e.g. 1-5°C) can also induce cell disorders that may progressively become more detrimental over time (Come, 1991). This physiological damage to plant tissues, brought about by extended exposure to low temperature, is commonly referred as chilling injury (CI). Depending on the species and the flower parts (e.g. petals, stem, leaves), the critical temperature that causes CI in cut flowers varies from 1 to about 5°C. In some cut flowers, including roses, many post-storage disorders, such as reduction in photosynthetic activity, flower wilting and discolouration, incomplete flower opening (Faragher *et al.*, 1986), and petal blueing (Leonard *et al.*, 2001) are partially related to chilling-induced alterations in metabolic processes.

Table 1.1: Production of the major cut flower species in Greece during sixteen years.

Source: Greek Ministry of Agriculture, (2004).

Flower species	**Production (10^6 single flower stems)**				
	1984	**1988**	**1992**	**1996**	**2000**
Carnations	234	230	218	202	185
Gladiolus	29	25	19	12	14
Chrysanthemum	16	35	27	31	34
Rose	41	50	66	71	69
Tulips	9	7	8	6	3
Gerbera	2	4	5	6	5

Table 1.2: Imports, exports and imports/exports ratio in million Greek drachmas of cut flowers grown in Greece from 1988 to 2000. Source: Greek Ministry of Agriculture (2004).

	Year				
	1988	**1992**	**1996**	**1998**	**2000**
Imports	1903	5394	8500	13026	12132
Exports	169	229	550	1071	980
Imports/Exports	11.2	23.5	15.4	12	12.4

1.2 AIMS AND OBJECTIVES

1.2.1 Overall aim

This study was carried out to investigate the effects of factors, such as season, storage conditions and vase solutions on postharvest behaviour of 'First Red' and 'Akito' roses grown in Crete. The practical aim was to study if 'First Red' and 'Akito' roses can be exported from the island of Crete all year round.

1.2.2 Objectives

- To study the effects of pre-harvest environmental changes on vase life of 'First Red' and 'Akito' roses grown in Crete.

There is considerable variation in the postharvest quality of cut flowers during the growing season. For example, roses grown during summer had longer vase life than flowers cut from plants grown during winter (Moe, 1975). This shortening of vase life of cut roses grown during winter depends on cultivar and may be due to early flowering (e.g. petal abscission, bent neck) and leaf status problems (e.g. wilting, drying) (Slootweg *et al.*, 2001). Temperature and relative humidity changes during the year can affect longevity and postharvest quality of cut roses (Mortensen and Fjeld, 1995; Mortensen and Fjeld, 1998; Shin *et al.*, 2001). The study, therefore, of possible seasonal variation in postharvest behaviour of cut roses grown in Crete

would have practical implications for cut flower industry in terms of exports and selling time. In this research, the regulation of different vase life parameters (e.g. vase life duration, chlorophyll fluorescence, transpiration rates, flower opening, flower colour) by environmental factors, such as temperature, relative humidity and light intensity, have been examined for two commercial rose cultivars which differ in flower colour (white and red petal colour). The influences of environmental conditions on response of flowers to low temperature storage (e.g. chilling tolerance) and different ABA treatments (e.g. pulse, spray and vase solution) were also studied.

- To investigate storage effects on vase life of roses.

Cold storage (wet or dry) at 0-10°C is the most effective method of maintaining the quality of cut rose flowers (Mayak and Faragher, 1986). Cold storage allows accumulation of sufficient stock for commercial consignment when the supplies are limited and regulates market supply when there is surplus production (Joyce *et al.*, 2000). Postharvest storage characteristics (e.g. temperature) are important with respect to marketing of cut flowers. The study of storage temperature for 'First Red' and 'Akito' roses grown in Crete may assist the cut flower industry of the island to reduce storage-related problems and promote exports.

- To study the effects of abscisic acid (ABA) and ABA-analogues, applied both before and after storage, on vase life of 'First red' and 'Akito' roses.

As mentioned above, low temperature storage may be harmful for cut flowers. Although abscisic acid (ABA) is a senescence hormone of plants, it can induce stomatal closure and extend vase life (Pompodakis and Joyce, 2003). Additionally, ABA has been found to regulate the plant response to cold stress possibly by inducing stomatal closure (Janowiak *et al.*, 1996; Ristic *et al.*, 1998; Janowiak *et al.*, 2002). When chilling sensitive plants, such as maize seedlings, are exposed to chilling temperatures, their endogenous ABA level increases (Janowiak *et al.*, 2002). Moreover, application of ABA to several sensitive species induced chilling tolerance (Pardossi *et al.*, 1992; Prasad *et al.*, 1994). Recently, ABA analogues, which are less expensive than pure ABA, have been found to reduce water loss in cut roses to the same degree as ABA (Pompodakis and Joyce, 2003). Thus, addition of ABA or ABA

analogues to vase water of cut roses before or after storage might be beneficial for preventing chilling injury (CI) and extending vase life.

1.3 THESIS STUCTURE

Pre-harvest effects on cut flower characteristics, storage and water relations of cut flowers and ABA are reviewed in the literature of Chapter 2. The materials and methods of experiments are described in Chapter 3. The effects of seasonal variation and storage temperature on vase life parameters of roses were examined in Chapter 4. The relationship between environment conditions in glasshouse and vase life characteristics was studied by measuring both pre-harvest growing conditions in glasshouse and vase life parameters in the laboratory. In Chapter 5, autumn and winter grown roses, which were most sensitive to low temperature storage, were examined. Different ABA treatments were applied to protect cut roses against low temperature damage. In the same chapter, histology studies of bent neck symptom are also presented. The effects of ABA and its analogues on Low Temperature Injury (LTI) of cut roses are presented in Chapter 6. Finally, in Chapter 7, overall conclusions based on the results of this study and opportunities for future work are discussed.

CHAPTER 2

LITERATURE REVIEW

2.1 PRE-HARVEST EFFECTS ON VASE LIFE

The appearance, quality, and longevity of cut flowers depend upon the conditions of cultivation, the correct harvest time and postharvest handlings. Flowers cultivated under optimal conditions exhibit the highest quality. 30-70% of the final quality of many cut flowers is predetermined at harvest (Halevy and Mayak, 1979). Factors, such as genotype and environmental conditions (temperature, relative humidity, atmospheric composition, nutrient solution, irrigation and substrate) seem to be most important for final flower quality (Halevy and Mayak, 1979). These factors can affect physical characteristics (diameter, weight, length), chemical composition (sugar content, anthocyanin and chlorophyll intensities), respiration, transpiration, and vase life of cut flowers (Celikel and Karacaly, 1995).

2.1.1 Genetic effects

The genotype of a rose cultivar can affect vase life of cut flowers. Any genes involved in the natural senescence process and water stress that cut flowers are subjected to during cultivation and after cutting can affect longevity and flower quality. For example, genotypic variation in stomatal closure and thus maintenance of water status in subsequent vase life can be seen for two cut rose cultivars (Mayak *et al.*, 1974). 'Golden Wave' rose flowers wilted very early and had a short vase life due to poor stomatal closure of leaves under water stress conditions. In contrast, 'Baccara' roses, which have a long vase life, are able to close their stomata to a greater degree, thus, reducing transpiration rate and water stress (Mayak *et al.*, 1974). In miniature roses, differences in vase life were due to differences in the expression of *RhETR*, a putative ethylene receptor gene. The expression of *RhETR* was distinctly higher in 'Bronze' with short flower life than in the longer life 'Vanilla' (Muller *et al.*, 2000).

Genotypes with different morphological and anatomical leaf characteristics require different conditions during cultivation (Mortensen, 2001). For example, 'Grand Prix' roses are suitable for low light growth conditions, but 'Tineke' roses are non-suitable. This may be due to the higher total chlorophyll *a* content in 'Grand Prix' than to 'Tineke' roses even at low light conditions (Sevelius *et al.*, 2001). Furthermore, leaves of *Rosa bracteata* were thinner than those of *Rosa rugosa* and had greater chlorophyll content per unit fresh weight and leaf area due to genetic differences (Ueda *et al.*, 2000). Some rose cultivars, such as 'Frisco', 'Golden Gate', 'Dream', and 'Kardinal' were moderately affected by high air humidity, while cultivars such as 'Orange Unique', 'Miracle', 'Prophyta', 'Amadeus' and others were more sensitive (Mortensen, 2001). Vase life of such sensitive cultivars was reduced due to increased rates of water loss after harvest.

2.1.2 Environmental conditions

Yield and quality of cut roses is determined by not only variety selection but also environmental conditions in which the plant is grown (Sancho, 1989). Seasonal changes during cultivation often influence the postharvest life of cut flowers (Halevy and Mayak, 1979; Torre *et al.*, 2001). Roses are generally more sensitive than other species to changes in climate conditions including lower irradiation level or increased temperature (Moe, 1975). Factors such as temperature, light, relative humidity and atmospheric CO_2 concentration in greenhouses can affect yield and quality of cut flowers (Torre *et al.*, 2001; Slootweg *et al.*, 2001).

Postharvest life can range from a few days to 1-2 weeks during the winter (Mortensen and Fjeld, 1995). Winter night temperatures lower than 0°C can decrease yield and quality of flowers. Lighting period and light intensity is also limited during winter. This results in lower photosynthetic rates (Sancho, 1989). Simultaneously, net photosynthesis decreases when rose plants are subjected to water stress. This is a specific problem in greenhouse roses grown particularly in summer. High temperatures, light intensity and low RH (%) cause excessive water loss from both plant and soil. This leads to increased water deficit stress. Maximum production can be expected with supplementary light and CO_2 in greenhouses along with a good control of irrigation, fertilisation, temperature, and air humidity (Sancho, 1989; Kramer, 1981).

2.1.2.1 Temperature

Both low (<18°C) and high (>25°C) temperatures a few days before harvest can reduce vase life of cut flowers. Several problems have been reported for different cut flower species (Table 2.1). Excessively high temperature during cultivation decreases vase life and quality of flowers. High temperatures reduce carbohydrate levels shortening vase life of freesias, tulips, irises and carnation flowers (Halevy and Mayak, 1979). The optimal day and night temperature in greenhouse rose production is generally 21°C and 18°C, respectively (Sancho, 1989). Cut 'Garnette' roses grown at 21°C had greater vase life as compared to roses grown at 15, 18, 24, and 27°C (Moe, 1975). However, water uptake increased by 10 ml with increase in growing temperature from 15 to 27°C. This resulted in a higher incidence of roses with bent neck (Moe, 1975). Plants of *Rosa rugosa* and *Rosa bracteata* showed maximum photosynthetic rates at 15°C and 25°C, respectively (Ueda *et al.*, 2000). The longevity of 'Zorina', 'Garnette', and 'Baccara' roses increased by more than 3 days as growing temperature increased from 12 to 24°C (Moe, 1975). Similarly, vase life of Bouvardia flowers was higher at growing temperature of 25°C than at 22, 18 and 15°C (van Gorsel, 1993). Transpiration rate was higher for Bouvardias grown at 15 or 18°C than at 22 or 25°C (van Gorsel, 1993). The mean production of 20 carnation cultivars was highest during the autumn and late spring, when the mean temperature was about 30°C (Lipari and Romano, 1989). Vase life of cut 'Astor' carnations also appeared higher during the autumn as compared to flowers grown the winter, spring and summer, respectively (Celikel and Karacaly, 1995).

The best quality of rose stems in terms of length, diameter and leaf are obtained at 18°C (Moe and Kristoffersen, 1969; Shin *et al.*, 2001). Flower stem length of 'Carl Red', 'Samantha', and 'Carinella' roses was shorter at 30°C than at 20°C. Conversely, leaf number per stem was increased when grown at 30°C (Yamaguchi and Hirata, 1998). Stem length, diameter and leaf area of 'Kardinal' roses generally decreased as mean air temperature increased from 15°C to 30°C. The stem diameter was related to pith diameter that influenced the accumulation and translocation of stored substances like starch and sugars (Shin *et al.*, 2001). Mean flower dry weight (D.W.) at 15°C was 3 g while at temperatures above 24°C it was > 2 g (Shin *et al.*, 2001). This may be due to increased carbohydrate consumption by

the respiration at higher temperatures (Shin *et al.*, 2001). Moe and Kristoffersen (1969) attributed the decrease in flower dry weight to high evapo-transpiration rate. The decrease of flower dry weight at high temperature is sometimes related to fewer and smaller petals. At low growing temperature (<15°C), the incomplete conversion of chloroplasts to chromoplasts can be responsible for the development of greenish tints on roses (Moe and Kristoffersen, 1969).

Table 2.1: Problems associated with growing temperature in different cut flower species. Data was obtained after harvest.

Species	**Temperature (°C)**	**Effects**	**Reference**
Freesia	30	E_1	Halevy and Mayak, 1979
Tulips	30	E_1	as above
Iris	30	E_1	as above
Carnations	30	E_1	as above
Chrysanthemum			
cv. Seiun	30	E_1	Adachi *et al.*, 2000
Rosa hybrida			
cv. Carl Red	30	E_2	Yamaguchi and Hirata, 1998
Samantha	30	E_2	as above
Carinella	30	E_2	as above
Baccara	30	E_3	Biran and Halevy, 1974a, b
Sonia	<15	E_4	Moe, 1988
Bouvardia			
cv. Bridesmaid	15-18	E_5	van Gorsel, 1993
Roxanne	15-18	E_5	as above
van Zijverden	15-18	E_5	as above

Note. E_1 = decreased sugar contents, E_2 = short stems, E_3 = petal blueing, E_4 = 'blackening' or 'greening' of petals, and E_5 = high transpiration.

2.1.2.2 Relative humidity

High relative humidity (90% as compared to 60%) during growth can cause high incidence of bent neck and leaf drying, decreasing vase life of cut flowers (Moe and Kristoffersen, 1969; Mortensen and Fjeld, 1998; Slootweg *et al.*, 2001). High relative humidity (91%) reduced the vase life of both 'First Red' and 'Golden Gate' roses (Mortensen and Gislerod, 1999). High air humidity slightly affected vase life of 'Frisco', 'Golden Gate', 'Dream' and 'Kardinal' roses, while it reduced the vase life of 'Orange Unique', 'Miracle', 'Prophyta' and 'Amadeus' (Mortensen, 2001). When these flowers were grown at high RH for more than 6 h daily, the flowers did not open properly after harvest. The air humidity around rose plants can be affected by the density of the plant canopy and the air movement around the leaves. Air humidity increases with greater canopy crowding than in the free air above the canopy (Mortensen, 2001).

The effects of RH during growth depend on the development stage of the plant. Three or four weeks at 90% RH in the last part of the growing period (flowering) significantly ($P < 0.05$) decreased the vase life of 'Souvenir' and 'Baronesse' roses as compared to 75 or 85 % RH through out the growing period (Mortensen and Fjeld, 1995). The water loss of detached leaves was also high (more than 50%) in leaves which had developed at 90% RH. Conversely, two or three weeks at 90% RH in the first part of the growth period followed by 65% RH, did not increase the incidence of bent neck or leaf drying as compared to constant 65% RH (Mortensen and Fjeld, 1995). Water loss of detached leaves was also increased for 14 rose cultivars grown at 91% RH as compared to 75 and 83%. Stem fresh weight decreased by 11% as the RH increased from 83 to 91% RH (Mortensen and Gislerod, 1999). High air humidity during cultivation increased the stem length of 'Souvenir' roses by 2-4 cm but it did not affect stem fresh weight (Mortensen and Fjeld, 1998). The effects of air humidity on time until flowering usually vary from species to species. For example, high air humidity did not affect flowering in cut roses and *Begonia* species, but did delay flowering in chrysanthemum. In *Saintpaulia ionantha* and *Campanula isophylla*, on the other hand, high air humidity promoted flowering (Mortensen *et al.*, 2001).

2.1.2.3 Light

Plants grown in shade have small leaves and bigger cell size (Sevelius *et al.*, 2001). Leaves of these plants are thinner and richer in chlorophyll (Boardman, 1977; Ueda *et al.*, 2000). Shading can cause a drastic decrease in ethylene production in the stems (Zieslin and Mor, 1990). Such morphological and physiological differences during cultivation can affect the quality and post-harvest life of cut flowers.

The yield and the quality of greenhouse roses generally increase with supplementary lighting mainly during winter because of the poor light conditions in Northern Europe. In Southern Europe (e.g. Crete, Greece), on the other hand, supplementary lighting is not required. High levels of supplementary lighting (150-200 μmol m^{-2} s^{-1} photosynthetic photon flux) are generally used in greenhouse rose production in Scandinavia, and very high yields are obtained during the dark winter period (Mortensen and Fjeld, 1998). Lighting period and light intensity are the most important characteristics of the supplementary light in the greenhouses.

Lighting period

Periodical lighting with night breaks in between cultivation induces photosynthesis more than continuous light. This may be due to sugar metabolism and translocation from the leaves to other plant parts (e.g. flowers), since carbohydrates are important for the longevity of cut roses (van Doorn *et al.*, 1991b; Sarkka *et al.*, 2001). Vase life of cut 'Frisco' roses was higher in treatments with two night breaks in lighting than treatments with a continuous night break (Sarkka *et al.*, 2001). Increasing the lighting period from 18 to 24 h, increased the number of flowering stems of different rose cultivars by 12% as a mean for all cultivars (Mortensen and Gislerod, 1999). However, water loss of detached leaves was also increased (Mortensen and Gislerod, 1999). Supplemental light disturbs the normal behaviour of the stomata, increasing the transpiration rates of cut flowers (Slootweg and van Meeteren, 1991). Thus, the incidence of bent neck and crisped leaves increased, leading to short vase life (Mortensen and Gislerod, 1999). Vase life of cut 'Souvenir' roses grown for large lighting periods (more than 16 h) was reduced. This was due to high incidence of bent neck and leaf drying (Mortensen and Fjeld, 1998).

Light intensity

Light intensity directly influences the efficiency of photosynthesis that determines the carbohydrate contents of flowers. Flowers containing relatively high amounts of carbohydrates, especially mobile sugars, last longer in the vase (Nowak and Rudnicki, 1990). Change in light intensity can alter leaf photosynthetic rate (Sevelius *et al.*, 2001). Net photosynthetic rates in mature leaves of *Rosa bracteata* and *Rosa rugosa* considerably increased as photosynthetic photon flux densities (PPFD) increased from 500 to 1000 μmol m^{-2} s^{-1}. Leaves of both rose species showed their highest photosynthetic rates at 1000 μmol m^{-2} s^{-1}, while greater light intensities (>1000 μmol m^{-2} s^{-1}) did not alter photosynthetic rates (Ueda *et al.*, 2000).

Supplemental light up to 370 μmol m^{-2} s^{-1} for 20 h per day increased rose yield and flower stem weight in the months from April to June in Norway (Mortensen *et al.*, 1992). Rose yield and rose quality were improved when 'Frisco' roses were grown with supplemental light between 3 and 174 μmol m^{-2} s^{-1}. Roses receiving supplementary light also produced more petals than traditional grown roses. When 'Gabriella' roses were grown in supplemental light (100 μmol m^{-2} s^{-1}), petal number per flower increased by *ca.* 1.15-fold (Bredmose, 1997). The number of rose stems during 10 month experimental period, as a mean for five rose cultivars ('Frisco', 'Kiss', 'Madelon', 'Cardinal' and 'Jaguar') increased by *ca.* 1.18, 1.41 and 1.53-fold whilst PPFD increased from 130 to 190, 250 and 370 μmol m^{-2} s^{-1}, respectively. At these light densities, from the period beginning of April until mid September, number of rose stems increased by 1.44- and 1.47-fold in cvs. 'Baronesse' and 'Kiss', respectively (Gislerod and Mortensen, 1997). Stem and root growth of 'Mercedes' roses was much higher at 180 μmol m^{-2} s^{-1} than at 90 μmol m^{-2} s^{-1} (Maas and Bakx, 1997). Moreover, supplementary PPFD at 0-174 μmol m^{-2} s^{-1}, increased fresh weight and dry weight of cut 'Gabriella' roses harvested in the spring (Bredmose, 1997).

Low light intensities and the short days of winter, on the other hand, decrease the longevity of cut carnations, gerberas, and roses in comparison with the warm, long days of summer (Nowak and Rudnicki, 1990). Low light intensity causes excessive elongation of flower stems and delays stem hardening. The insufficient hardening (lignification) commonly results in bent neck of certain roses and stem-bending in carnations and gerberas (Burdett, 1970; Nowak and Rudnicki, 1990). Excessive shading of roses during the formations of anthocyanins in petals produces petal

blueing. Roses grown under low light intensities produced petals with normal colour when treated with a sugar solution during opening, while petals left on the plants had a paler petal colour (Halevy and Mayak, 1974). This indicates that colour intensity of petals is affected by carbohydrate concentration in surrounding tissues that is low in flowers grown in low light intensities. Excessive light intensity is also unfavourable to flower quality. Light in large amounts can produce red colouration of tissues, leaf spots, browning, and leaf drop (Nowak and Rudnicki, 1990). Recently, some rose varieties have been reported as suitable for low light conditions after measurements of relative chlorophyll fluorescence (F_v/F_m) on the leaves (Sevelius *et al.*, 2001). F_v/F_m values of 'Grand Prix' and 'Tineke' roses were higher when the plants were grown at low light intensity (80 μmol m^{-2} s^{-1}) as compared to roses grown at high light intensity (200 μmol m^{-2} s^{-1}). The lower F_v/F_m of both varieties under high light intensities indicates a developed stress and susceptibility to photoinhibition (Sevelius *et al.*, 2001).

2.1.2.4 Carbon dioxide (CO_2)

Increasing CO_2 concentration in greenhouses can increase plant tolerance to water deficit stress by improving the water utilisation (Sancho, 1989; Wittwer, 1983). Increasing CO_2 concentration can act to close leaf stomata reducing transpiration rates. This is desirable in areas of high water stress like the Mediterranean. However, if stomata are already closed, CO_2 will have little effect (Kramer, 1981). CO_2 can also be beneficial in light-deficient areas and/or seasons (Wittwer, 1983). CO_2 can also act as inhibitor of ethylene synthesis. In carnation and *Ipomoea* flowers, CO_2 delayed the development of in-rolling of petal tissue which was related to ethylene production (Halevy and Mayak, 1979).

Yield and quality of rose flowers grown in greenhouses have been increased by maintaining the CO_2 concentration around 800 μmol mol^{-1} (Mortensen, 2001). CO_2 enrichment can improve whole plant net CO_2 exchange rate (NCER) of miniature roses (Jiao *et al.*, 1990). However, NCER efficiency can be variety dependent. For example, although NCER of 'Orange Sunblaze' and 'Sunbird' roses increased as the CO_2 concentration rose from 300 to 1300 μl l^{-1}, 'Orange Sunblaze' roses showed significantly higher NCER than 'Sunbird' roses (Jiao *et al.*, 1990).

Increased CO_2 can increase photosynthesis efficiency and production rates especially in conditions of low light intensities (Sancho, 1989). An increase in concentration of atmospheric CO_2 from 330 to 1000 μl l^{-1} increased photosynthetic rate (Wittwer, 1983). The rate of photosynthesis measured at frequent intervals on single leaves of plants grown in 660 μl l^{-1} of CO_2, increased 1.41-fold for soybean, 1.15-fold for cotton, 1.07-fold for sunflower and 0.2-fold for sorghum above plants grown at 330 μl l^{-1} (Kramer, 1981). Accordingly, fresh weight and stem elongation increases under conditions with supplementary CO_2 in the greenhouses (Kramer, 1981). Increasing the CO_2 level during rose flower development at low light intensities increased pigmentation and decreased blueing (Halevy and Mayak, 1979). The time of flowering is also affected by increased CO_2 concentrations. Floral initiation was delayed at 1000 μl l^{-1} of atmospheric CO_2 in sorghum, corn, sunflowers and cotton (Kramer, 1981).

However, concentrations of atmospheric CO_2 more than 1000 μl l^{-1} can reduce photosynthetic rates. High CO_2 can cause leaf wilting by accumulation of starch in leaves and deformation of chloroplasts (Kramer, 1981). Starch concentration increased in both leaves and stems of 'Meijikatar' miniature roses as the CO_2 concentration increased from 350 to 1050 μl l^{-1} after 28 days of exposure. CO_2 enrichment also increased both sucrose and glucose concentrations in the leaves of 'Meijikatar' miniature roses but it did not affect fructose levels. In contrast, carbohydrate status in flower stem did not change by increasing CO_2 concentration (Rajapakse *et al.*, 1997).

2.1.3 Irrigation

Water stress at the stage of flowering has been reported to be crucial for a range of crops. Flower-bud differentiation, particularly during sexual organ formation, is moisture-sensitive in most annual and perennial crops (Salter and Goode, 1967). In roses, water shortage at these stages can result in defective floral organs, sterility, reduced number of flowers and failure to open (Chimonidou-Pavlidou, 1999).

Water stress delayed the final marketable stage and decreased the size of petals of 'Madelon' roses when it occurs at early stages of flower development (leaf primordia formation) (Chimonidou-Pavlidou, 1999). Although, water stress at early

stages caused a delay in the production cycle, it did not have negative effects on the quality of flower stems (stem length, fresh weight, bud dimensions and number of petals). However, water stress at the stage prior to petal initiation reduced incidence of well-formed petals and the height of the flower buds (Chimonidou-Pavlidou, 1999). Gladioli cv. Sans Souci were also sensitive at a very early stage when flower initiation took place and later at the fourth leaf stage through to flower stem elongation (Halevy, 1972). On the other hand, carnations grown under a low irrigation regime, which induced severe water stress, can produce flowers with better vase life than those grown at wet and normal irrigation regimes (Halevy and Mayak, 1979).

Flower buds appeared earlier and flower stems were shorter when rose 'Madelon' were irrigated once a week than every two days (Chimonidou-Pavlidou, 2001). The acceleration of bud appearance and the shorter stem length may be due to the protection mechanisms of rose plants under water stress conditions (Chimonidou-Pavlidou, 2001).

2.1.4 Fertilisation

In cut flower production, management of fertilisation is important in ensuring the high productivity. Fertilization is important for growth, flowering and vase life of cut flowers (Roychowdhury and Roychowdhury, 1995). Adequate mineral nutrition is pre-requisite to obtaining large increases in yield from enhancement in the greenhouses (Kramer, 1981).

2.1.4.1 Nitrogen (N)

Under normal growing conditions, with sufficient light as a source of energy, plants use nitrogen to form proteins. Enzyme systems in green parts of plants rapidly reduce nitrate-N (NO_3^-) to intermediate compounds that are subsequently converted into amino-nitrogen. Organic acids arise from carbohydrate metabolism in combination with the amino-nitrogen to yield amino acid (Cabrera, 2001). In greenhouses, nitrogen is applied in irrigation water at concentrations of 150-250 mg l^{-1} mainly in the form of nitrate (NO_3^-) (Cabrera, 2001). The average N absorbed by plants of 'Garden Giant' narcissus was about 400 mg $plant^{-1}$ including original N (i.e.

N already in the plant before additional N is taken up) and additional N (Ruamrungsri *et al.*, 1997). Maximal yields were obtained with 90 mg l^{-1} N in commercially grown roses. Yields increased in 'Royalty' roses grown with N up to 60 mg l^{-1} but they showed a decrease at 220 mg l^{-1} (Cabrera *et al.*, 1993; Cabrera, 2000). Ammonium (NH_4^+) is not often used, as it is generally associated with toxicity problems (Cabrera, 2001). Nitrate produced chrysanthemum flowers with better longevity than ammonium or urea (Halevy and Mayak, 1979). Cabrera (2001) did not find significant effects of NO_3^-/NH_4^+ ratio on yield and quality of 'Bridal Pink' roses. However, 75:25 NO_3^-/NH_4^+ ratio has been reported as beneficial nutrient solution for roses (Feigin *et al.*, 1986).

Nitrogen deficiency limits the formation of new tissue, thus, can reduce photosynthetic rate (Kramer, 1981). Nitrogen deficiency symptoms, such as leaf chlorosis, reduce rose quality (Cabrera, 2000). High nitrogen concentrations in nutrient solution can cause toxicity symptoms after harvest in some flowers (Table 2.2). In chrysanthemum, high nitrogen at the later part of the growing period decreases the postharvest quality of the flowers (Halevy and Mayak, 1979). Simultaneously, total soluble carbohydrates also decreased as the nitrogen concentration increased from 50 to 200 mg l^{-1} in chrysanthemum plants (Druege *et al.*, 2000). Concentrations of non-structural carbohydrates like sugar and starch and the sucrose:starch ratio in leaves usually decrease with increasing nitrogen supply (Druege, 2001). Excessive nitrogen fertilisation can also increase susceptibility to infection, that is, grey mould (*Botrytis cinerea*) during storage.

2.1.4.2 Phosphorus (P)

Phosphorus (P) is a component of adenosine triphosphate (ATP). ATP serves as the major energy source within the cell to produce a number of biological processes such as photosynthesis and the synthesis of proteins (Taiz and Zeiger, 1998). P deficiency can decrease the number of flower buds in petunia and the growth parameters (fresh weight, height, plant diameter, shoot number, leaf size and root evaluation) in Osteospermum (Nowak, 2001). P can accumulate to toxic levels when applied as frequently as nitrogen and potassium.

2.1.4.3 Potassium (K^+)

K^+ activates many enzymes, controls the level of sugars and plays a role in photosynthesis and in translocation of photosynthates (De La Guardia and Benlloch, 1980). K^+ application in the field showed positive effects on quality and vase life of gladiolus spikes. This may be due to improved water balance brought about by accelerated enzyme activity (Roychowdhury and Roychowdhury, 1995). K^+ deficient plants have thinner walls, smaller xylem vessels, and are more sensitive to water stress (Halevy and Mayak, 1979). In roses, bent neck incidence increases at low K^+ concentration in the foliage (Johansson, 1979). However, high K^+ concentration can enhance pedicel elongation and increase the tendency for the plants at high RH to develop dry spots on the leaves. Extracellular K^+ can cause weakness in the structure of the plasma membrane by removing calcium (Ca^{2+}) from the outer surface of the plasma membrane (Torre *et al.*, 2001).

2.1.4.4 Calcium (Ca^{2+})

Ca^{2+} plays an important role in horticultural practice, due to its effects on plant structure and metabolism (Baas and Marissen, 2000). Increased Ca^{2+} content in plant tissue can inhibit the development of some diseases after harvest by preventing the activity of polygalacturonase and other pectic enzymes (Gislerod, 1999). The degradation of pectins which leads to deterioration of tissues is also mediated by polygalacturonase activity (Baas and Marissen, 2000). The structure of root system can also be affected by Ca^{2+} concentration. At the low Ca^{2+} levels (0.62 mM l^{-1}), the roots of 'Mercedes' roses were short, thick and with few branches but at the higher levels (2.5 and 5.0 mM l^{-1}), a fibrous root system developed. $Ca(NO_3)_2$ also reduced stem softness and the bending of carnation stems (Halevy *et al.*, 1978).

High RH can reduce Ca^{2+} content of both flowers and leaves in rose plants by decreasing transpiration rates (Torre *et al.*, 2001). The increase in Ca^{2+} content of leaves and flowers by adding Ca^{2+} in the nutrient solution was more effective at moderate RH than high RH (Torre *et al.*, 2001). In roses, removal of part of the leaves and/or bent-down stems increased Ca concentration in the remaining leaves and petals (Baas *et al.*, 2003). Both K^+ and Ca^{2+} concentrations in the flower stem seem to influence the post-harvest life of pot roses. Increased keeping life in pot roses

was caused by decrease in K^+: Ca^{2+} ratio. A decrease in flower wilting and *B. cinerea* infection was also found as Ca^{2+} concentration increased (Mortensen *et al.*, 2001).

2.1.4.5 Other minerals

Magnesium, copper, zinc and boron may become deficient in certain soils. Deficiency of these elements can cause a wide variety of symptoms before and after harvest, depending on the flower species (Halevy and Mayak, 1979). Boron is used in sugar translocation, synthesis of nucleic acids, starch formation, and carbohydrate synthesis. Iron is used in chorophyll synthesis and in electron transfer processes. Manganese and copper are used to activate several enzymes for nitrogen metabolism and synthesis. Deficiencies of boron can decrease carnation longevity (Halevy and Mayak, 1979). Symptoms of manganese or zinc deficiencies can appear if pesticides (e.g. Mancozeb) containing these elements are not used regularly.

2.1.4.6 Salinity

Soluble salts increase osmotic potential. Thus, water in saline soils is less available to plants. This generally leads to physiological water stress. Increased concentration of ions, mainly Cl^- and Na^+ can lead to saline toxicity (Wahome *et al.*, 2001). Roses are sensitive to salinity. Rose production is reduced when soil solution electrical conductivity (EC) exceeds 2 to 3 mS cm^{-1} and Na^+ and Cl^- concentrations are higher than 1-4 mM (Hughes and Hanan, 1978). In *Anthurium*, EC at 1 mS cm^{-1} is sufficient for the production of flowers, but 1.2 mS cm^{-1} seems to be more favourable for the flower quality (Dufour and Clairon, 1997).

The damage in the older foliage and plants of both 'Bridal Pink' and 'Grenoble' roses receiving high NaCl application was only caused by presence of Cl^-, not Na^+ (Cabrera, 2001). In *Rosa chinensis* and *R. rubiginosa*, NaCl caused necrosis initially on the lower older leaves leading to eventual abscission. Cl^- may be responsible for the leaf necrosis, since Cl^- concentration is higher than Na^+ concentration especially in the upper and lower leaves (Wahome *et al.*, 2001). High soil salinity reduced chrysanthemum flower longevity especially under infrequent irrigation because of increased water stress (Halevy and Mayak, 1979). Salt stress of

roots can also raise abscisic acid (ABA) concentration in leaves, roots and xylem sap (Druege, 2001).

2.1.5 Stage of development at harvest

Flowers maintain a fresh appearance much longer if they are harvested at the appropriate stage of development. The commercial development stage of the flower at harvest varies in different flower species (Appendix 2, Table A2.1) and is affected by environmental conditions, the distance from market and the consumer preference. Flowers are generally cut at the bud stage, when they are closed with good turgor in the petals (Halevy and Mayak, 1979). Some flower species, such as carnations, roses, chrysanthemums, etc., can be harvested at less advanced stages in summer than in winter in order to develop properly in the vase (Nowak and Rudnicki, 1990). However, some flower species are not able to open if they are cut at an early stage. Moreover, the incidence of bent neck for roses and gerberas is high in flowers cut too early. This is due to insufficient lignification of the vascular tissue in the neck region at early development stages (Kohl, 1961; van Meeteren, 1978). Flowers can be stored longer, when they are cut at early stage of development (bud stage) if chemicals are used to promote opening (Faragher *et al.*, 1984).

2.1.6 Time of harvest

The time of harvest is another important parameter that can drastically affect vase life. Morning cutting has advantages in terms of better turgidity. Morning harvest is recommended for flowers that lose water quickly after cutting, as in the case of roses (Nowak and Rudnicki, 1990). Flowers cut late in the afternoon, on the other hand, last longer than those cut in the morning, indicating higher levels of carbohydrates in the flower stem during the afternoon (Halevy and Mayak, 1979). This can be observed in flowers bearing leaves, such as roses, but not in flowers having scapes, such as gerbera (Halevy and Mayak, 1979). Harvesting at high temperature and high light intensity should be avoided since flowers are at low turgor.

Table 2.2: Effects of nitrogen concentration in the nutrient solution on post-harvest life of ornamental products.

Plant Species	Product	Maximum level (mmol N l^{-1})	Symptoms	Reference
Euphorbia pulcherrrima	potted plant	21.4	leaf drop	Ter Hell and Hendriks 1995
Impatiens New-Guinea	potted plant	10.7	bud drop	as above
Rosa hybrida	potted plant	14.3	as above	as above
Campanula carpatica	potted plant	14.6	decreased flower life	Serek, 1990
Cyclamen persicum	potted plant	38.7 [A]	bud drop, leaf senescence	Druege, 2001
Rosa hybrids	cut flower	21.4	decreased vase life	Menard *et al.*, 1995
Cyclamen persicum	cut flower	11.4	no effect	Druege, 2001
Sandersonia aurantiaca	cut flower	42.9	no effect	Clark and Burge 1999
Cytromium sp.	cut flower	321 [B]	no effect	Druege, 2001
Pteris sp.	cut flower	321 [B]	increased longevity	as above
Dendranthema grandiflora	cutting	14.3	storage ability	Druege *et al.*, 1998

[A] =concentration in the substrate; [B] = mmol N $plant^{-1}$ $year^{-1}$

2.2 STORAGE OF CUT FLOWERS

Suitable postharvest storage and transport procedures of cut flowers are required for maintenance of flower quality during transport from the production site to the market place (Miranda *et al.*, 2000). Several problems are associated with storage of cut flowers that affect vase life (Table 2.3). Flowers may look fresh after storage but they do not last as long as fresh flowers in the vase. Failure of buds to open during vase life is a major problem in roses, narcissus, chrysanthemums, and some iris cultivars (Halevy and Mayak, 1981). At high storage temperature, failure opening to acceptable commercial stage is also a common problem during vase life. In red roses and carnations, petal discoloration often occurs, decreasing flower quality. Foliage discolouration (e.g. yellowing) is also a problem in some leafy flowers after storage (Halevy and Mayak, 1981). Storage characteristics, such as temperature and duration, considerably affect quality and longevity of cut flowers.

2.2.1 Storage temperature

Many cut flowers are stored at low temperature in order to reduce respiration and transpiration rates and thus prolong longevity. The action of ethylene is limited for roses stored at low storage temperature, although ethylene biosynthesis follows a climacteric course (Mayak and Faragher, 1986). The benefit of low temperature during storage is not only to reduce the rate of metabolism of flower stems, but also that of microorganisms in the stems. The increase in bacterial load of cut 'Sonia' roses during dry storage at 5°C was less than during dry storage at 20°C (van Doorn and De Witte, 1991a).

Cut 'Bridal Pink' roses stored at 5°C retained better quality and had longer vase life than those stored at 20°C (Hu *et al.*, 1998b). Vase life of 'Mercedes' roses also decreased as storage temperature was raised from 0 to 8°C. The decrease of vase life in higher storage temperature was attributed to increased ethylene production. The rate of ethylene production in 'Mercedes' roses stored at 0 and 3°C rose to a maximum during the first two days of vase life and then declined (Faragher *et al.*, 1986). Kangaroo paw 'Bush Dawn' flowers stored at 13°C for 1 - 4 weeks showed highest rate of reduction in vase life, as compared to flowers at 0 and 7.5°C, respectively (Miranda *et al.*, 2000). These flowers showed the minimum rate of

reduction in vase life, when they were stored at 7.5°C. Vase life of *Grevillea* 'Sylvia' inflorescences stored at 0°C, which did not differ from the vase life of non-stored inflorescences, was higher as compared to inflorescences stored at 5 and 10°C, respectively (Joyce *et al.*, 2000).

2.2.1.1 Chilling injury

Chilling injury (CI) is a physiological disorder induced by low but, not freezing, temperatures (Marangoni *et al.*, 1996). At low temperatures, the membrane lipids are more rigid ('bulk lipid-phase membrane transitions'), resulting in a redistribution of the proteins (Come, 1991). This phenomenon induces a modification of membrane permeability and decrease of enzyme activities (Come, 1991). About 2-5% of the total lipid is believed to undergo these phase transitions both in chloroplasts and mitochondria (Raison and Orr, 1986). A main effect of CI is the damage of plasma and thylakoid membranes in chilling sensitive plant tissues resulting in the rapid chloroplast deterioration (Ristic *et al.*, 1998). The modification of membrane structure brought about by low temperatures can induce cell disorders, thus causing injuries. Sensitive plant tissues show a variety of symptoms, often after being transferred to non-chilling temperatures (Marangoni *et al.*, 1996).

Platt-Aloia and Thomson (1987) suggested the existence of chilling-induced lateral phase separation (LPS) in the plasma membrane of avocado fruit. LPS can alter the biophysical properties of membranes, which are related to their composition, and thereby affecting functionality (Marangoni *et al.*, 1996). LPS may be reversible depending on the point in time where lipid degradation and accumulation of lipid degradation products induce irreversible membrane damage (Figure 2.1) (Marangoni *et al.*, 1996). At this early stage, post-chilling recovery may be possible if the threshold of irreversibility has not been reached. Accordingly, when maize (*Zea mays* L.) seedlings, which are sensitive to CI, were returned to 25°C after a 5-d chilling period at 5°C, leaves quickly recovered photosynthetic activity (Aroca *et al.*, 2001; Janowiak *et al.*, 2002), indicating that the possible LPS was reversible. In some cases, this recovery was very pronounced in tolerant genotypes, reaching values that were even higher than the values before the chilling stress was administered (Janowiak *et al.*, 2002).

Low temperature induces oxidative stress in the cell. Under aerobic conditions, superoxide radicals and H_2O_2 are found to be normal metabolites (Prasad *et al.*, 1994). These metabolites are kept at low, steady-state levels by the action of antioxidant enzymes, such as superoxide dismutase and CAT (Spychalla and Desborough, 1990), glutathione reductase (Hausladen and Alscher, 1994), superoxide dismutase and dehydroascorbate peroxidase (Sen Gupta *et al.*, 1993) and ascorbate peroxidase (Prasad *et al.*, 1994). These enzymes are mainly located in the organelles and cytosol (Prasad *et al.*, 1994).

One of the main causes of CI is the production of reactive oxygen species during chilling, which are greatly increased in light induced photooxidation (Aroca *et al.*, 2001). Reactive oxygen species are produced during chilling conditions because enzyme activities of Calvin-Benson cycle are delayed. Under these conditions, the electron transport pathway is restricted leading to excess energy absorption by oxygen (Aroca *et al.*, 2001). According to Wise (1995), there are three main mechanisms to diminish photo-oxidation during chilling: a) avoiding production of reactive oxygen species by diminishing electron transport chain, b) dissipating excess energy as heat via violoxanthin de-epoxidation and c) scavenging reactive oxygen species formed by antioxidant compound and enzymes.

Chilling injury effects on cut flowers

Low temperatures sometimes induce cell disorders that may become detrimental to some flowers. Flowers originated from tropical or subtropical climates are most sensitive to CI and require storage at 7-15°C to prevent CI symptoms (Nowak and Rudnicki, 1990). The susceptibility of these flowers to CI is often due to increased proportion of unsaturated lipids in plasma membrane (Marangoni *et al.*, 1996). The severity of CI depends on the cut flower species, flower maturity, the temperature applied, the period of exposure and the duration of cold treatment (Nowak and Rudnicki, 1990). The typical CI symptoms of cut flowers, such as petal fading and/or blueing (Leonard *et al.*, 2001), necrotic lesions on petals and leaves or foliage wilting (Hu *et al.*, 1998a, b) and delay in bud development after storage (Faragher *et al.*, 1986), are often visible in the first days of vase life. Kangaroo paw stems that suffered CI collapsed in the region immediately below the flower (Joyce and Shorter, 2000). Long term low temperature storage reduced flower opening of

both 'H1' and 'Bush Dawn' flowers. 'H1' and 'Bush Dawn' kangaroo paw flowers stored at 0°C showed signs of wilting and discolouration earlier than flowers at 2.5, 5 and 7.5°C (Joyce and Shorter, 2000).

An associated cause of CI is photooxidation caused by photoinhibition associated with low temperatures (Krause and Weis, 1984). Photoinhibition can act as a photo-protective mechanism when excess absorbed excitation energy is dissipated as heat within the PSII antennae by zeaxanthin-related quenching. The electron transport pathway from the Q_A (the primary electron acceptor of PSII) to cytochromes, through plastoquinone molecules, is blocked in low temperatures (Krause and Weis, 1984). This decreases the quantum efficiency for photosynthesis determined by gas exchange, and decreases the variable to maximum fluorescence yield (F_v/F_m) ratio (Boese *et al.*, 1997). F_v/F_m of kangaroo paw 'Bush Dawn' flowers stored at 7.5°C was the highest. F_v/F_m value for flowers stored at 0°C was the lowest, indicating CI effects on structure and efficiency of photosystem II (Miranda *et al.*, 2000). Hakam *et al.* (2000) also reported decrease in F_v of roses with decreasing temperature from 25 to 0°C followed a pattern similar to that of other crops, e.g. *Arachis hypogaea* L., *Zea mays* L. and *Oryza sativa* L., that are susceptible to CI (Smillie and Hetherington, 1990).

2.2.2 Storage duration

Cut flowers are perishable commodities that were traditionally grown close to the centres of consumption. However, long distance transport is increasingly required by cut flower industry (Hu *et al.*, 1998b). Thereby, cut flowers may be stored for short or long periods. Duration of storage varies from species to species depending on the distance from the market (Hu *et al.*, 1998a, b; Miranda *et al.*, 2000; Joyce and Shorter 2000). Some flowers, such as *Astible*, *Bouvardia*, *Gypsophila*, *Alchemilla*, *Celosia*, *Gaillardia*, *Godetia*, *Hesperis*, *Limonium*, *Molucella*, *Nigella*, *Ornithogalum*, and *Salvia* are sensitive to storage. Other species, such as *Lilium*, *Tulipa*, *Iris*, and *Dianthus*, can be stored dry for long periods without significant reduction in water uptake (van Doorn, 1997).

The maximum storage period for roses has been reported to be 1 or 2 weeks at 0-5°C (Del Rio *et al.*, 1989). The quality of cut 'Sonia' and 'Baccara' rose stems kept for 3 weeks at 1-2°C was unacceptable because of high incidence of *B. cinerea*

infection (Del Rio *et al.*, 1989). 'Madame Delbard' roses were able to tolerate storage more than 6 days without major effects but vase life was higher when flower stems were not stored. Vase life of 'First Red' roses was reduced after a storage period of 6 days due to disease problems. The incidence of diseases also increased while vase life and flower opening decreased in 'Classy' roses increasing the storage duration from 3 to 12 days (Leonard *et al.*, 2001). Cut 'Visa' roses did not show Botrytis infection during the first week of storage (Serrano *et al.*, 1992). After 2 weeks the percentage of flowers with infections increased progressively as the storage duration increased from 2 to 7 weeks (Serrano *et al.*, 1992). Carnations can be stored at 4°C for 2 weeks but longer periods of cold storage lead to shorter vase life. Ethylene production was higher and occurred earlier in 'White-Sim' carnations stored at 4°C for 5, 7 and 9 weeks than flowers stored for 2 weeks (Serrano *et al.*, 1995).

Both vase life and F_v/F_m value for kangaroo paw 'Bush Dawn' cut flowers progressively declined as storage duration increased from 1 to 4 weeks (Miranda *et al.*, 2000). Storage for 4 weeks also reduced vase life of 'H1' and 'Bush Dawn' kangaroo paw flowers compared to non-stored control flowers (Joyce and Shorter, 2000). Storage period more than 6 days significantly decreased the vase life of *Grevillea* 'Sylvia' inflorescences. This was due to flower abscission caused by increased ethylene production (Joyce *et al.*, 2000). Sucrose concentrations in cut chrysanthemums increased during the first two weeks of storage but after that period the values declined during the last two weeks (Druege *et al.*, 2000).

2.2.3 Wet storage

The wet storage method is used by cut flower industry for short term (1 to 3 days) storage. This method is used for cultivars that are not suitable for long term storage. Flowers stored wet are normally harvested at a more advanced stage (Halevy and Mayak, 1981). When cut 'Bridal Pink' roses were stored wet with their stem bases in deionised water, they maintained their fresh weight for the first days of vase life. However, addition of bactericides and a carbohydrate in the water during wet storage may be more useful. Addition of 0.1 M fructose along with 0.3 mM 8-hydrixyquinoline significantly increased vase life, while sucrose and glucose had no significant effects (Hu *et al.*, 1998b).

Table 2.3. Some negative effects of storage (duration and temperature) on vase life of different cut flower species.

Flower species	Period	Temperature (°C)	Method	Effects	Reference
Kangaroo paw					
cv. Bush Dawn	1 – 4 [A]	0	dry	chilling injury	Miranda *et al.*, 2000
cv. Bush Fever	14 [B]	1	dry	flower wilting and/or fading	Jones and Faragher, 1991
Roses					
cv. Sonia	1 – 4 [B]	20	dry	increased bacterial counts	van Doorn and De Witte, 1991a
cv. Mercedes	3 and 8 [B]	8	dry	increased ethylene production	Faragher *et al.*, 1986
cv. Bridal Pink	1 – 3 [B]	5	wet	decreased water loss	Hu *et al.*, 1998a
cv. Bridal Pink	20 [B]	1	dry	50% bent necks	Hu *et al.*, 1998b
cv. Sonia, Baccara	21 [B]	1-2		*Botrytis cinerea* infection	Del Rio *et al.*, 1989
cv. Sonia	1 – 2 [B]	20	dry	increased transpiration after storage	Jeong-Seob *et al.*, 1992
cv. Visa	2 [C]	4	wet	increased activity of ACC-synthetase	Serrano *et al.*, 1992
cv. Mercedes	10 [B]	2	dry	decreased flower opening	Faragher *et al.*, 1984
Carnations					
cv. White-Sim	4 [A]	4	wet	increased activity of ACC-synthetase	Serrano *et al.*, 1995

Note. [A] = weeks; [B] = days; [C] = hours.

STRESS

(i.e., LOW TEMPERATURE)

↓

MEMBRANE BIOPHYSICAL CHANGES

REVERSIBLE LATERAL PHASE SEPERATION ARE EVIDENT

↓

INDUCTION OF COMPOSITIONAL (CHEMICAL) ALTERATIONS IN

MEMBRANE LIPIDS (CATABOLIC AND ANABOLIC CHANGES)

↓

IRREVERSIBLE LATERAL PHASE SEPERATIONS

(DUE TO ACCUMULATION OF LIPID DEGRADATION PRODUCTS)

↓

CHILLING INJURY SYMPTOMS

(ONLY DUE TO IRREVERSIBLE EFFECTS)

Figure 2.1: Sequence of metabolic events leading from a stress-induced alteration in the properties of membranes to observable macroscopic tissue damage (after Marangoni *et al.*, 1996).

2.2.4 Dry storage

Cut flowers are stored dry after harvest for long periods (>1 week) in order to be fresh during transportation. Carnations and narcissus are better stored dry in the tight bud stage and then opened in a proper opening solution while roses, chrysanthemums and iris stored in the bud stage usually fail to open properly and to reach the desired quality after storage. Other flowers, such as gladiolus or *Strelitzia*, should be pulsed before storage. At the end of dry storage, flower stems should be re-cut and pulsed in a solution (Halevy and Mayak, 1981). Sucrose and bactericides at concentrations higher than those in vase solutions are mainly used as pulse treatment after dry storage. The optimum concentration depends on the cut flower species. For example, sucrose is used at 20% or higher for gladiolus, gerbera, and *Eremurus*, 10% for carnations, *Strelitzia*, and *Gypsophila*, 2-5% for roses, chrysanthemum, and *Leucospermum* (Halevy and Mayak, 1981). In contrast to the suggestion of Halevy and Mayak (1981), sugar at >2% caused necrotic spots (leaf crisping) on the leaves of cut 'Kardinal' roses due to induced high tissue osmotic potentials (Markhart and Harper, 1995). Sugar accumulated in the apoplast and caused leaf crisping (Markhart and Harper, 1995). The use of a commercial preservative solution containing sugar caused serious damage on leaves of roses after just 1 day in the vase life room (Torre and Fjeld, 2001). In cut 'Baccara' roses, provision of sucrose in the vase solition at 4% gave advanced leaf crisping, resulting in much reduced flower stem quality (Pompodakis and Joyce, 2003). Most sensitive to leaf crisping are rose stems grown in closed greenhouses with supplemental light, elevated CO_2, and high relative humidity. Such flowers have a high transpiration rate (absorption) after harvest and more sucrose from the vase solution accumulates during vase life, thereby causing leaf damage (Markhart and Harper, 1995; Torre and Fjeld, 2001; Pompodakis and Joyce, 2003).

After dry storage, transpiration of 'Sonia' roses was progressively reduced, indicating an occlusion in the stems (Jeong-Seob *et al.*, 1992). After 72 hours dry storage at 5°C, cut 'Bridal Pink' roses lost about 20% of their fresh weight, their leaf and petal water potentials were low and all flower stems had bent neck problems (Hu *et al.*, 1998a). The water potential of petals was higher than leaf water potential, indicating that water moves from the petals to the transpiring leaves during dry storage (Hu *et al.*, 1998a). When cut roses were stored wet at 20°C, water loss was

low and the flower stems did not show bent neck problems (Hu *et al.*, 1998b). Although the water content in petals of cut 'Mercedes' roses increased during wet storage, the vase life of flowers stored wet reduced as compared to flowers stored dry (Faragher *et al.*, 1986). Grevillea 'Sylvia' inflorescences stored wet at 22°C for 2 days showed a higher increase (17.0%) in fresh weight compared to the increase (2.8) during dry storage at 22°C for 2 days (Joyce *et al.*, 2000). However, inflorescences had longer vase life after dry storage than after wet storage. Decreased vase life was attributed to higher metabolism during wet storage.

2.2.5 Ethylene

Ethylene is generally produced in many flowers in three phases. In flower buds and young flowers, production is low and stable. During flower maturation, ethylene production sharply increases reaching a maximum level. Afterwards, ethylene decreases and again remains stable at a low level (Nowak and Redneck, 1990). Senescence of carnation flowers is associated with a climacteric-like increase in ethylene production (Serrano *et al.*, 2001). Similarly, ethylene production by petals of intact and cut rose flowers initially remains constant at a low level for a few days and then increases sharply until flowers senescence (Halevy and Mayak, 1975).

Enrichement of ethylene in the atmosphere accelerates the autocatalytic production of ethylene by flowers. Autocatalytic production of ethylene is due to increased membrane permeability of tonoplast (the membrane separating vacuole from cytoplasm) that is caused by exogenous ethylene. Increase in membrane permeability of tonoplast causes the translocation of ethylene precursors from the vacuole to the cytoplasmatic membranes or cytoplasm where enzymes are synthesizing ethylene (Nowak and Rudnicki, 1990). Exogenous ethylene reduces the longevity of roses, but it is not known whether ethylene is a natural regulator of senescence in roses. Ethylene did not appear to be an important natural regulator of the postharvest life of roses. Ethylene is involved in the postharvest life of cut roses only when it is present as an atmospheric pollutant, or when its synthesis is stimulated by stress (Reid *et al.*, 1989a, b). The substrate for ethylene production in plant tissue is methionine, the amino acid containing sulphur. In the chain of biochemical reactions, methionine converts to *S*-adenosylmethionine (SAM). During the climacteric of some cut flowers, there is an increase in the activities of 1-

aminocyclopropane-1-carboxylic acid synthase (ACC synthase) and ACC oxidase (Serrano *et al.*, 2001). ACC synthase and oxidace are enzymes that catalyse the conversion of *S*-adenosylmethionine (SAM) to ACC and ACC to ethylene, respectively (Figure 2.2).

Progressing senescence of flowers increases their sensitivity to ethylene. Some flowers that are highly susceptible to ethylene are sensitive to concentrations as low as 1-3 μl l^{-1} during 24 h exposure. Conversely, less sensitive flowers are resistant to concentrations 10-100 times higher (Nowak and Rudnicki, 1990). Increased ethylene concentration can cause chlorophyll breakdown and colour fading which are characteristic features of leaf and flower senescence, respectively (Table 2.4). Exogenous ethylene was reported to induce deterioration of cell membranes in carnations (Eze *et al.*, 1986). In many potted roses, relatively low concentrations of ethylene (0.1, 0.5, or 1.0 μl l^{-1}) caused considerable reduction in display quality and in longevity, indicating the need for chemical protection from ethylene action (Muller *et al.*, 1998). In many members of the Geraniaceae such as *Pelargonium* species, continuous exposure to 1 μl l^{-1} ethylene causes petal abscission reducing the visual impact of the potted plant (Cameron and Reid, 2001). Moreover, flower abscission in Geraldton waxflower (*Chamelaucium uncinatum*) was caused by exogenous ethylene (Joyce, 1993). Abscised flowers can increase the incidence of *Botrytis spp.* infection and other saprophytic pathogens. Infection of waxflower by *B. cinerea* can enhance ethylene production leading to unacceptable levels of flower abscission (Joyce, 1993; Tomas *et al.*, 1995; Terry *et al.*, 2003). During pollination, *Pelargonium* flower petals also abscise even without exposure to exogenous ethylene (Cameron and Reid, 2001). In *Iris* flowers, on the other hand, tepal senescence was not regulated by ethylene (van Doorn *et al.*, 1995).

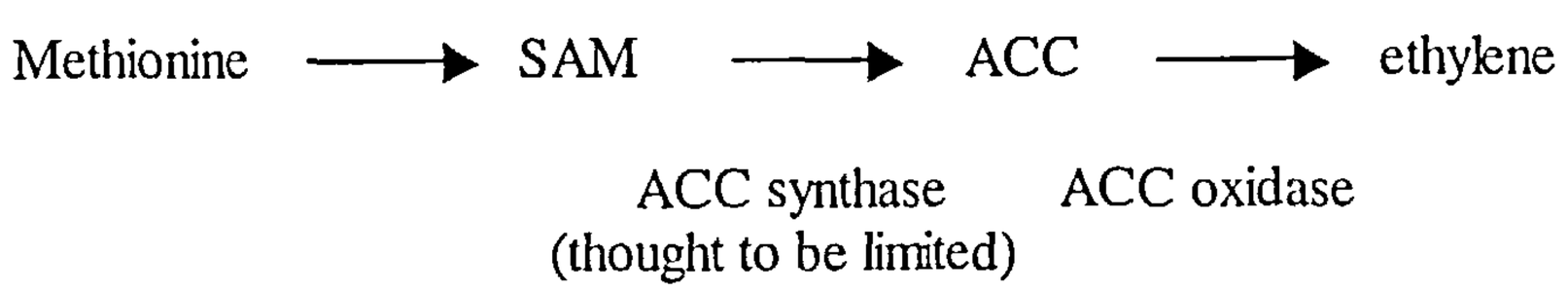


Figure 2.2: The process of ethylene synthesis.

Table 2.4: Symptoms of ethylene toxicity in selected cut flower species.

Plant name	Toxicity symptoms	Reference
Alstromeria peregrina	flower malformation, petal darkening, petal abscission	van Doorn, 2001
Camellia japonica	petal abscission, necrotic browning of petals and stamens	Doi and Reid, 1996
Ceratopetalum gummiferum	floral organ abscission, decreased vase life	Macnish *et al.*, 2000b
Chamelaucium uncinatum	floral organ abscission	Joyce, 1988; 1989; Macnish *et al.*, 2000a,b
Chrysanthemum	slight hastening of senescence	Nowak and Rudnicki, 1990
Dianthus caryophyllus	bud sleepiness, petal wilting	Borochov *et al.*, 1982
Euphorbia (scarlet plume)	leaf yellowing and abscission	Nowak and Rudnicki, 1990
Eustoma grandiflorum	hastening of senescence	Ichimura *et al.*, 1998
Freesia	bud malformation or blasting, hastening of senescence	Spikman, 1986; Nowak and Rudnicki, 1990
Gerbera	slight hastening of senescence	Nowak and Rudnicki, 1990
Grevillea	floral organ abscission, decreased vase life	Macnish *et al.*, 2000a,b
Helianthus maximilianii, Penstemon digitalis, Weigela sp.	decreased vase life	Redman *et al.*, 2002
Iris (bulbous)	bud sleepiness or blasting, hastening of senescence	as above
Lathyrus odoratus	floral abscission	Sexton *et al.*, 1995
Leptospermum petersonii	floral organ abscission, decreased vase life	Macnish *et al.*, 2000b

Table 2.4: Continued.

Plant name	Toxicity symptoms	Reference
Lilium longiflorum	bud sleepiness or blasting, petal abscission, inhibition of bud opening, green lowermost flower buds	Elgar *et al.*, 1999
Narcissus (daffodil)	smaller flower diameters, hastening of senescence	Nowak and Rudnicki, 1990
Nerine	petal blueing, hastening of senescence	as above
Orchids (*Cattleya, Dendrobium, Phalaenopsis, Paphiopedilum, Vanda*)	reddish flower colour, epinasty, hastening of senescence	as above
Phlox paniculata	flower abscission, inhibition of flower opening	Porat *et al.*, 1995
Rosa hybrida	inhibition of bud opening, petal epinasty, petal blueing, hastening of senescence	Goszczynska and Zieslin, 1993; Muller *et al.*, 2001
Snapdragon	floret abscission	Nowak and Rudnicki, 1990
Tulipa gesneriana	bud sleepiness, petal blueing, hastening of senescence	as above
Verticordia nitens	floral organ abscission, decreased vase life	Macnish *et al.*, 2000b

2.2.5.1 Ethylene inhibitors

Ethylene can be removed from the cold storage rooms simply by ventilating with fresh air. It can also be removed by using air scrubbers containing the solution of potassium permanganate ($KMnO_4$) that oxidizes ethylene. Use of ethylene scrubbers, however, is not recommended because excessive amounts of the materials are required to control ethylene levels (Halevy and Mayak, 1981). UV light or X-ray radiation may also be used to clear the air of ethylene, but these methods are not widely used. The production of ethylene and its effects on flowers are also inhibited by CO_2. Increasing the CO_2 content in a cold room prevents the harmful effects of ethylene but may also cause injuries to some sensitive flowers (Nowak and Rudnicki, 1990).

Silver thiosulphate (STS) acts as an ethylene antagonist, since Ag^+ is able to travel throughout the cut flower (Knee, 1995; van Doorn *et al.*, 1991d). However, the mechanism of the effect of STS on ethylene action is still unclear (Ohkawa *et al.*, 1999). It seems that the action of Ag^+ is very specific since the other metal ions cannot act as Ag^+ (Taiz and Zeiger, 1998). Addition of 0.5 mM STS to the vase water increased the vase life of both fresh and stored 'Mercedes' roses by 2 days, reducing the rates of ethylene production (Faragher *et al.*, 1986). Pre-treatment of 'Lovely Girl' roses with between 0.5 and 1 μmol STS before exposure to 0.5 μl^{-1} l ethylene improved flower opening (Reid *et al.*, 1989a). STS at 0.1 mM for 24 hours significantly extended vase life and inhibited ethylene production from petals of *Eustoma grandiflorum* flowers (Ichimura *et al.*, 1998). In potted flowering plants, such as *Campanula carpatica* (bellflower) and *Schlumbergera truncata* (Christmas cactus), STS also inhibited the deleterious effects of exogenous ethylene (Serek and Sisler, 2001).

During the last 10 years, 1-Methylcyclopropene (1-MCP) has been used as an ethylene inhibitor in flowers. In miniature roses, 1-MCP is an important tool that can increase postharvest life after ethylene exposure for ethylene-sensitive rose cultivars, such as 'Bronze' and 'Charming'. In contrast, 1-MCP may be of little value to ethylene-insensitive cultivars, such as 'Vanilla' (Muller *et al.*, 1998). In several flowering plants (*Begonia elatior*, *Rosa hybrida* cv. Victory Parade, and *Kalanchoe blossfeldiana* cv. Tropicana), 1-MCP at 20 nl l^{-1} (6 h) provided as much protection as 0.5 mM STS, preventing ethylene-associated phenomena, such as flowers abscission,

leaf abscission, and flowers senescence (Serek *et al.*, 1994). 1-MCP at 10 nl l^{-1} prevented floral organ absission and associated loss in vase life in native Australian cut flower species, such as *Ceratopetalum gummiferum*, *Chamelaucium uncinatum*, *Grevillea* 'Kay Williams', *Grevillea* 'Misty Pink', *Leptospermum petersonii*, and *Verticordia nitens*, following treatment with 10 µl l^{-1} exogenous ethylene (Macnish *et al.*, 2000b). In *Campanula carpatica*, 1-MCP at 20 nl l^{-1} for 6 h prevented flower senescence and extended plant life by reducing ethylene amount. However, higher concentrations did not give additional improvement in flower longevity or plant display life (Serek and Sisler, 2001). A 6-h treatmnent with 20 nl l^{-1} 1-MCP completely inhibited the ethylene-induced flower absission of *Phlox paniculata* flowers (Porat *et al.*, 1995). In *Schlumbergera truncate*, on the other hand, 1-MCP at 50 and 100 nl l^{-1} extended plant display life and improved flower longevity, respectively (Serek and Sisler, 2001). Exposure of 1 µl l^{-1} 1-MCP completely inhibited petal abscission of *Pelargonium peltatum* that caused by increased ethylene levels (Cameron and Reid, 2001). When *Lilium longiflorum* (cv. Cordelia) flowers were pre-treated with STS or 1-MCP before exposure to ethylene had decreased ACC oxidase activity compared to those without pre-treatments (Elgar *et al.*, 1999).

2.2.6 Other storage parameters

Other parameters during storage including RH, light and CO_2 concentration can affect longevity and quality of cut flowers. Humidity control is important during storage. RH should be maintained as high as possible (90-95%) without causing free water during storage of cut flowers (Halevy and Mayak, 1981). Free water can increase disease incidence of grey mould caused by *B. cinerea*. *B. cinerea* disease severity of three rose cultivars markedly reduced, when flower stems were stored at reduced humidity levels between 50 and 80% (Hammer and Marois, 1989). Lighting in storage has little or no effect on longevity of cut flowers except chrysanthemum, dahlia and pyrethrum where light can cause leaf yellowing (Halevy and Mayak, 1981).

Controlled atmosphere (CA) storage has been proposed for long-term storage of cut flowers (Rogers, 1973). CA is applied to prevent ethylene producton and reduce transpiration rates of ornamentals. Longevity of some flowers, including roses is maintained by providing reduced oxygen and increased CO_2 levels in the storage

room at 1-2°C which decrease respiration rates and ethylene effects. Cut flowers can also be stored in chambers under low pressure (LP). Carnations stored up to 79 days retained 0.8-fold their original vase life comparing to non-stored control flowers (Dilley, 1977). Ethylene and CO_2 production rates were reduced and/or delayed after 5 weeks for carnations stored under LP compared to non-stored control flowers (Dilley, 1977). Staby *et al.* (1984) also showed that LP storage can extend vase lives of roses and carnations. However, the benefits were greater when the LP storage was combinined with pre- and post-storage treatments with STS.

2.3 WATER RELATIONS OF CUT FLOWERS

Water uptake and transport, water loss, and the capacity of the flower tissue to retain water are all components of water balance of cut flowers (Halevy and Mayak, 1981). These physicochemical processes are correlated with one another and with vase life. Maintenance of an optimal balance between vase solution uptake and transpiration is key to extending the vase life of cut flowers (Dixon and Peterson, 1989). Microbial growth, pH, plant growth regulator chemicals, sucrose and other carbohydrates, leaf removal, darkness, and mineral salts have all been shown to affect the water balance of the cut flower (van Doorn, 1997). Fresh weight of cut flowers declines, when water loss exceeds water uptake (for instance, under low RH conditions) (Rogers, 1973). Typically, cut flowers initially increase and subsequently decrease in fresh weight during vase life (Rogers, 1973). In roses, for example, loss of petal turgidity and fresh weight was preceded by a decreased rate of water uptake (Burdett, 1970).

2.3.1 Water uptake

The rate of water uptake of cut flowers depends, among other factors, on transpirational pull, the environment conditions, such as temperature, RH and light period and intensity, during vase life and the composition of vase solution. From the first to third day after harvest, water uptake exceeds water loss increasing cut flower weight (Carpenter and Rasmussen, 1974). The rate of uptake will then reach steady state corresponding to the rate of transpiration (van Doorn, 1997). This can vary among different species. In *Rosa*, *Bouvardia*, *Astilbe*, *Zantedeschia*, some

Chrysanthemum cultivars, *Polianthes tuberosa*, and several species from Australia (such as Kangaroo paw, *Chamelaucium*, *Banksia*, *Grevillea*, *Thrypyomene*, *Leptospermum*, and *Telopea*) the rate of water uptake rapidly declines after cutting (Mayak *et al.*, 1974). In other cut flowers, such as *Heliconia*, there is little water uptake even shortly after cutting and placement in water (Donselman and Broschat, 1986). In *Iris* flowers, water uptake decreased before tepal wilting and was reduced more by transpiration. Accordingly, water balance became negative before tepal wilting (van Doorn *et al.*, 1995). Water uptake of cut 'Sonia' roses, which showed symptoms of petal wilting and bent neck, was >10% of initial water uptake (De Stigter, 1980).

2.3.2 Water loss

Transpiration rate declines but tends to be higher than the water uptake rate during vase life (De Stigter, 1980). This difference results in a negative water balance (equals to the rate of uptake - rate of transpiration), a decrease in tissue water potential, and stomatal closure (De Stigter, 1980). Both flowers and leaves lose turgor and show signs of wilting, when the rate of water uptake is lower than the rate of transpiration (van Doorn, 1997).

Cut flowers lose water from all organs (i.e. leaves, stems, flowers) (Halevy and Mayak, 1981). Cut flowers with relatively small leaf area, such as carnations, lose much less water per stem over time than those with relatively high leaf area, such as lilies or roses (van Doorn, 1997). Water loss in cut flowers occurs much more rapidly through open stomata than through the cuticle (van Doorn, 1997). Only about 5% of the water loss escapes trough the cuticle. Almost all water lost from typical leaves occurs by diffusion of water vapour through stomata (Taiz and Zeiger, 1998).

2.3.2.1 Stomatal transpiration

Environmental conditions, such as light intensity and quality, temperature, relative humidity, can affect the activity of guard cells, regulating stomatal opening (Taiz and Zeiger, 1998). For example, if leaves kept in the dark are suddenly illuminated, light is received by guard cells as a signal for stomatal opening (Taiz and Zeiger, 1998). The presence of functional or non-functional stomata and the reaction

of stomata to developing water stress is also important (van Doorn, 1997). Stomata are usually present in all green epidermal tissues and sometimes in the epidermis of non-green parts, such as petals. In some species, stomata are present on the stamens of flowers. The number of stomata in green tissues differs from species to species (Table 2.5). Stomatal opening in cut flowers is often delayed after a period of reduced water supply (van Doorn, 1997).

Table 2.5: Presence of stomata on the petals of some cut flowers.

Genus	Cultivar	Number of stomata	Reference
Arand	Christine	38 A_1	Hew *et al.*, 1987
		40 A_1	
	Wendy Scott	45 A_1	Hew *et al.*, 1987
		45 A_1	
Cymbidium	Alexalban; Sirius;	None	
	Tapestry King Arthur	Some. A_1 only	
Dendranthema	Variety not known	Some	
	Reagan; Cassa	20 A_2 only	
Dianthus caryophyllus	White Sim; Scania	None	
Gerbera	Mickey;	None	
	Liesbeth; Tamara		
Lilium	Enchantment	10 A_2 only	
Rosa	Lady Seton;	None	Stubbs and Francis, 1971
	Golden Wave;	None	Mayak and Halevy, 1974
	Sonia; Madelon; Ilona;	None	
	Motrea; Frisco; Cara Mia	None	
Tulipa	Apeldoorn; Frappant	500 A_2	
		100 A_1	
		100 A_2	
		10 A_1	

Note. Data published by van Doorn (1997), unless otherwise indicated. A_1 = Abaxial side (underside of petal); A_2 = Adaxial side (upperside of petal).

2.3.2.2 Cuticular transpiration

Even after stomatal closure, water loss may still occur in some flowers via flower stem (van Doorn, 1997). For example, in *Astilbe*, with numerous small flowers, the leaves and stem accounted for only 40% of total water loss. The rate of transpiration may depend on thickness of cuticle (van Doorn, 1997). Rose stems lack stomata and have a limited effect (> 5%) on the water loss via flower stem. Carnation stems, on the other hand, have large numbers of stomata and account for 40% of water loss (Carpenter and Rasmussen, 1974).

2.3.3 Water stress

Cut flowers are exposed to developing water stress as a result of continuous water loss. This leads to increasingly limited water uptake after harvest (van Doorn, 1997). Water stress during the period in the vase is, therefore, often the cause of shortened vase life (Mayak *et al.*, 1985). Water stress can develop because of a stem blockage, but also as a function of excessive transpiration rate (van Doorn, 1997). Water stress can also cause stomatal closure, which reduces the rate of transpiration (section 2.4.4 for details in ABA action under water stress conditions). A temporary water stress caused a decline in protein synthesis enzymes and a decrease in protein content in *Iris germanica* flowers (Halevy and Mayak, 1981). These flowers recovered, and protein content was partially replaced after rehydration (Paulin, 1975). Temporary water stress did not, however, induce any change in carbohydrate (hexoses and sucrose) content in cut carnations (Halevy and Mayak, 1981). In green tissue, water stress has been shown to stimulate ethylene production (Apelbaum and Yang, 1981).

Because of stomatal closure during water stress, CO_2 concentration inside the leaf is limited, resulting in lower rate of photosynthesis during vase life (Schapendonk *et al.*, 1992; Kramer, 1981). In addition to stomatal closure, water stress may reduce the photosynthetic capacity directly, either by inhibiting the Calvin cycle or the electron transport rate over the chloroplast membranes (Schapendonk *et al.*, 1992). In contrast to the above hypothesis of lower electron flow under water stress, Flexas *et al.* (2000) found that electron transport rate of grapevine leaves under water stress was

still *ca.* 0.75-fold of control values. Thus, it was concluded that water stress did not alter greatly the electron transport rate.

2.3.4 Stem blockage

Premature loss of turgor and thus water deficit in many species of cut flowers, such as roses and chrysanthemums, is due to an occlusion in the water conducting system (van Doorn *et al.*, 1991c; 1995). Flowers of Zantedeschia aethiopica and Z. elliotiana, for example, decreased in fresh weight early in vase life. This loss was caused by occlusions in the xylem of the scapes (Tjia and Funnell, 1986). In cut roses, premature wilting and bent neck were found to be due to a blockage by bacteria in the basal stem segment (Mayak *et el.*, 1974). In Iris flowers, on the other hand, stem occlusions did not decrease water potential, which remained at about – 0.2 to – 0.3 MPa. Early tepal wilting in Iris was due to processes inherent in the tepal cells, such as ethylene-induced senescence (van Doorn *et al.*, 1995).

Blockage of cut flowers is usually present at the lower end of the stems, on the cut surface, and/or inside the stem-end xylem elements (van Doorn, 1997). Possible causes of vascular blockage are air emboli, micro-organisms and physiological wound healing processes in stems as a response to cutting (van Doorn *et al.*, 1989). Vascular blockage has also been caused by enzymatic activity or endogenous ethylene formation (Put and Rombouts, 1989). Blockage can be caused by bacterial growth inside the stems, but when microbial growth is excluded or kept to minimum a physiological occlusion was also observed (van Doorn and Otma, 1994).

2.3.4.1 Physical causes

The presence of air in stem vessels can disrupt water conductivity and, as a result, cause water deficit (van Doorn, 1995). Air entering the base of the cut stems during harvest and/or during storage may form emboli that seriously impair rehydration of cut flowers (Halevy and Mayak, 1981). Stems of 'Cara Mia' roses showed a maximum vascular occlusion after 3 hours of exposure to air (van Doorn and Otma, 1994). Xylem sap can also be disrupted by the development of gas bubbles (cavitation) at near-vacuum tensions (van Doorn, 1995). Cavitation may be a cause of the vascular blockage that develops during dry storage (van Doorn and Suiro, 1996).

Vessels with larger diameter cavitate earlier than those with a small diameter (van Doorn and Suiro, 1996). Using counts of ultrasonic acoustic emissions, numerous cavitations were detected in *Thryptomene saxicola* stems (van Doorn and Jones, 1994). The presence of cavitated xylem elements was also observed in 'Samantha' roses (Dixon and Peterson, 1989). In 'Cara Mia' roses, the rate of water uptake was correlated with the presence of cavitated elements (and possibly tracheids) in the stems (van Doorn and Suiro, 1996). In chrysanthemum flowers, cavitations in the upper stem lead to air embolism and subsequent flower wilting. Wilting of florets was also due to physical blockage of xylem vessels in the stem base (Singh and Moore, 1992).

2.3.4.2 Microbiological causes

Microorganisms found in flower stem or were growth in vase water resulted in low hydraulic conductivity of the stems, especially in the basal stem segment (van Doorn and De Witte, 1991b). The developed occlusions in the stems of cut rose flowers have been correlated with increasing numbers of microorganisms in stems (Florack *et al.*, 1996). Postharvest increase in stem flow resistance was dependent on the presence of microorganisms in 'Forever Yours' roses (Marousky, 1969). High bacterial counts in stems were also correlated with vascular occlusion in the petioles of *Adiantum raddianum* fern fronds (van Doorn *et al.*, 1991a). Not only bacteria but also yeasts and filamentous fungi can lead to vascular blockage (Put and Clerkx, 1988).

Bacterial occlusions

Bacterial occlusions can cause interruption of water uptake and wilting problems in cut flowers. Solution turbidity attributable to bacterial growth in the vase was negatively correlated with both vase life ($P \leq 0.01$; $r^2 = -0.48$) and vase solution usage ($P \leq 0.05$; $r^2 = -0.65$) by cut 'Baccara' roses (Pompodakis *et al.*, 2004). A high number of bacteria were also detected using light microscopy at the cut surface of cut dahlia flowers (*Dahlia variabilis*), which had been placed in water for some days (van Doorn, 1997). Ultrastructural investigations of cut roses also showed that a

population of bacteria at the cut surface was responsible for vascular occlusion (van Doorn *et al.*, 1991c). In 'Sonia' roses held in water for seven days, the lowest hydraulic conductance in the base of the stems correlated with the highest number of bacteria (van Doorn *et al.*, 1989).

Reduction of vase life of 'Sonia' roses was found with bacterial inoculations of 10^6 cfu ml^{-1} (Put and Jansen, 1989). The vase life of 'White Sim' carnations was also reduced when the vase water was inoculated with $\leq 10^8$ cfu ml^{-1} (van Doorn *et al.*, 1995). Bacteria were not found in the vase water of cut roses, but a considerable number of bacteria were present in the stems after a few days of vase life. These bacteria were able to cause vascular occlusion within the flower stem (van Doorn and Perik, 1990). When vascular blockage developed, the bacterial population in the basal 5-cm stem segments was about 10^6 cfu gfw^{-1} (van Doorn, 1997). Numbers of bacteria in the vase water of gerbera flowers that induced a curvature of more than 90° were 10^6 and 10^8 colony forming units (cfu) ml^{-1} in the cultivars 'Liesbeth' and 'Mickey', respectively (van Doorn *et al.*, 1994). Bacterial counts reach a maximum of about 10^7 cfu ml^{-1} of water after a few days of vase life in roses, carnations, tulips, and chrysanthemums (van Doorn, 1997). When cut 'Sonia' roses were put in the vases for 1 to 4 days, the number of bacteria in the basal 5-cm of the stems was linearly correlated with the number of bacteria in the vase solution, indicating that bacteria in the flower stems contaminate the vase solution (van Doorn and De Witte, 1991a). Bacteria found in the vase water of many cut flower species, such as *Achromobacter*, *Alcaligenes*, *Bacillus*, *Escherichia*, *Flavobacterium*, *Micrococcus*, and *Pseudomonas* (Appendix 2, Table A2.2). In the vase water of roses, the predominant bacterium was *Pseudomonas*, while *Enterobacter* was a minor accompanying genus (Florack *et al.*, 1996). In vase water of carnations, about 50% of the bacteria were *Pseudomonas*, along with about 20% *Acinetobacter* and 20% *Alcaligenes* (van Doorn, 1997).

Action of Bacteria

The effect of bacteria on vase life is physical (van Doorn, 1997). Hydraulic conductivity of rose stems placed in either 5×10^9 or 2×10^7 cfu ml^{-1} of either living or dead bacteria declined rapidly. This was affected by temperatures at 1°C as at 20°C (De Witte and van Doorn, 1992). Occlusion by bacteria does not depend on their

physiological activity or that of the plant (van Doorn, 1997). Bacterial extracellular polysaccharides and globular proteins can result in vascular blockage (De witte and van Doorn, 1988). When bacterial growth was eliminated, vascular occlusion was still found, apparently due to the produced extracellular polysaccharides (van Doorn, 1997). Vascular blockage was also found in the basal end of the stems of cut roses when the extracellular polysaccharides from *Pseudomonas cepacia* were added to sterile water (De Witte and van Doorn, 1992). A thick layer of material consisting of bacteria with associated extracellular polysaccharides is often found to cover the xylem at the cut surface after a few days in the vase (van Doorn, 1997).

Microorganisms through their pectic enzymes may also impede the xylem cells, inducing numerous loose vessel fragments (Put and Rombouts, 1989). When pectolytic enzymes, such as pectate lyase produced by *Pseudomonas fluorescens* and polygalacturonase produced by *Kluyveromyces fragilis* were added to vase water of 'Sonia' roses, water relations were disturbed. This may be due to enzymatic degradation of the structures of xylem vessels (Put and Rombouts, 1989). However, van Doorn (1997) suggested that the bacteria isolated from vase water of cut rose flowers did not show pectolytic activity. The materials observed by Put and Rombouts (1989) were actually ice crystals, not wall fragments (van Doorn, 1997).

Some fungi species have also been found in the vase water of cut flowers (Appendix 2, Table A2.3). Fungi may have a role in the stem occlusion of some flowers, since captan (a fungicide) improved water uptake (van Doorn, 1997). Vascular blockage was observed when fungal extracellular polysaccharides were added into the vase water of cut rose flowers (Put and Klop, 1990).

Antimicrobial compounds

The use of antimicrobial compounds in the water, before storage, during wet-storage and during vase life has been found to extend vase life of cut flowers, such as roses, gerberas, gladioli, and antirrhinums (Hoogerwerf and van Doorn, 1992). Addition of some antimicrobial compounds to the vase water of cut flowers reduced the number of bacteria, which may be the reason for increased vase life (van Doorn *et al.*, 1990). Many different antimicrobial compounds at various concentrations are added into vase water of cut flowers (Appendix 2, Table A2.4). However, most

antimicrobial compounds, when used at high concentrations that adequately control microbial growth, are toxic to cut flowers (van Doorn, 1997).

Acidic solutions are also favourable for rehydration of cut flowers. Water flow increased through rose stem segments with decreasing pH from 6 to 3 (Durkin, 1979b). In cut 'Baccara' roses, decreasing solution pH from 8 to 6 enhanced flower water relations, fresh weight maintenance and vase life (Pompodakis *et al.*, 2004). The positive effect of low pH was attributed to reduction of microbial populations (Halevy and Mayak, 1979; Pompodakis *et al.*, 2004). Furthermore, low pH retarded stem blockage of roses in bacteria-free water (Marousky, 1971). Bacteria growth is initially suppressed at low pH, but a population of yeasts rapidly develops and many filamentous fungi are found (van Doorn, 1997).

2.3.4.3 Physiological causes

Physiological wound reactions in the stem may lead to occlusions in stems placed in water directly after harvest (Lineberger and Steponkus, 1976). Deposition of materials, such as suberin, lignin, tannin, or various gums were observed in the lumen of the xylem conduits of roses after cutting (van Doorn, 1997). Cutting may activate enzymes (e.g. peroxidases and phenylalanine ammonia lyase) involved in the biosynthesis of lignin and other substances that are deposited in cell walls or vessel lumens (Cline and Neely, 1983; van Doorn and Cruz, 2000). Some polysaccharide gums were found in the vase water of roses (Dixon and Peterson, 1989). Some species also exude latex at the cut surface after cutting. Latex consists of high molecular weight polyterpenes that are normally deposited in the vacuole (van Doorn, 1997). Tyloses are outgrowths of cell that a balloon structure in the lumina of the xylem conduits and may fill the conduit lumen. Tyloses are also found under the cut surface of cut flowers such as as *Prunus* spp., roses, and lilacs. Tyloses and latex generally originate from ray cells found in parenchyma cells (van Doorn, 1997).

Some chemicals have been used to prevent the latex blockage. Dipping the cut base in alcohol or holding the stem in boiling water can coagulate the latex (Halevy and Mayak, 1981). However, immersing the base of dahlia stems in boiling water is detrimental, while warm water at 50°C was slightly beneficial to vase life. In contrast, stem base dip in boiling water for 1 second in poinsettia and for 30 seconds for 'Iceland' poppy was beneficial (Halevy and Mayak, 1981).

2.4 ABSCISIC ACID

Abscisic acid (ABA) is a plant growth inhibitor that can have opposing roles during the vase life of cut flowers (Mayak and Halevy, 1972). ABA can induce stomatal closure in leaves, and, thereby, preventing transpiration. On the other hand, ABA can accelerate senescence, especially in leafless flower stems (Mayak and Halevy, 1972).

2.4.1 Changes in ABA levels of cut flowers

Endogenous levels of ABA in roses are generally greater at the end of vase life (during flower senescence) as compared to day 0 (Mayak and Halevy, 1972; Garello *et al.*, 1995). ABA contents of petals of cut rose flowers decreased during the first 3 days after harvest (Borochov *et al.*, 1976b; Le Page-Degivry *et al.*, 1991). A similar decrease occurred in isolated petals within the first 4 d of water stress (Le Page-Degivry *et al.*, 1991). Afterwards, however, ABA levels increased in petals, when senescence became clearly visible (Le Page-Degivry *et al.*, 1991; Muller *et al.*, 1999). In carnations, ABA contents in both petals and green tissue paralleled the ethylene climacteric rise in the petals (Hanley and Bramlage, 1989). ABA content in day lily (*Hemerocallis* hybrid) petals increased slowly from 24 h before flower opening and then more rapidly starting 12 h before opening (Panavas *et al.*, 1998; Rubinstein, 2000).

ABA levels in short-lived 'Dr. Verhage' roses were higher than that of long-lived 'Lovita' roses (Halevy and Mayak, 1975). In primary leaves of beans, Eze *et al.* (1981) suggested that the capacity to synthesize ABA was highest in young expanding tissues. A similar conclusion was also suggested by Eze *et al.* (1986) for carnation flowers. However, in roses an equal capacity of ABA synthesis for 1- and 8-day-old isolated petals was found by Le Page-Degivry *et al.* (1991). These opposing results may suggest that the capacity of plant tissues to synthesize ABA is not only dependent on their age but also on the plant species.

2.4.2 ABA-related flower senescence

Exogenous application of ABA often accelerates senescence of cut flowers (Barthe *et al.*, 1991). ABA has been found to induce a reduction in protein content in combination with increase in pH levels of the petal extract in roses (Borochov *et al.*, 1976a). In leafless flower stems, ABA when used at 10 mg l^{-1} promoted senescence of 'Golden Wave' and 'Super star' roses (Halevy *et al.*, 1974). In miniature potted roses cvs. Vanilla and Bronze, ABA treatment also promoted senescence in absence of leaves (Muller *et al.*, 1999). Vase life of roses was decreased and flower blueing was observed in the presence of 10^{-4} M of ABA in the vase water (Borochov *et al.*, 1976a). Application of 10^{-4} M ABA to cut petunia flowers accelerated their senescence and caused blueing in the petals (Vardi and Mayak, 1989). ABA enhanced senescence in darkness, when stomata had been closed (Halevy and Mayak, 1981).

2.4.2.1 Relationship of ABA and ethylene during senescence

A strong relationship between ABA and ethylene production has been reported in literature for cut flowers. Ethylene production levels were increased by ABA promoting senescence of cut carnations (Mayak and Dilley, 1976). ABA applications were also found to increase sensitivity of carnation flowers to ethylene (Borochov and Woodson, 1989). An increase in ABA levels of rose petals occurs several days after an increase in ethylene synthesis which may suggest that ethylene induces the rise in ABA levels (Mayak and Halevy, 1972; Halevy and Mayak, 1975; van Reinhold, 1991). Increase in ABA contents in petals of carnations, before the losses of fresh weight, was also associated with ethylene production (Borochov and Woodson, 1989).

In day lily (*Hemerocallis* hybrid), which is an example of ethylene-insensitive flower, ABA application accelerates senescence-associated changes in petals, such as increase in ion leakage, lipid peroxidation and activities of proteinases and nucleases (Panavas *et al.*, 1998; Rubinstein, 2000). Spraying miniature potted roses with 0.1 mM ABA clearly increased leaf yellowing and subsequent leaf drop by 6.6- and 3.4-fold in 'Bronze' and 'Vanilla' flowers, respectively (Muller *et al.*, 1999). In the same experiment with miniature roses, treatment of whole plants with the ethylene action inhibitor 1-MCP did not reduce ABA-induced leaf drop. These findings agree with

the observations of Zacarias and Reid (1990), who found that in Arabidopsis the promotion of senescence by ABA is not mediated through its stimulation of ethylene production.

2.4.3 ABA-induced stomatal closure

ABA supplied in the vase solution can induce stomatal closure in the leaves of cut flowers, including roses (Halevy *et al.*, 1974). Exogenous application with 10 mg l^{-1} of ABA in vase water of cut roses delayed flower fading (Halevy *et al.*, 1974). ABA at 1 mg l^{-1} in vase solution or at 10 mg l^{-1} for a 1 day pulse also delayed wilting and extended longevity of cut roses, apparently via stomatal closure (Kohl and Rundle, 1972). In cut 'Baccara' roses, addition of 10^{-5} M ABA or PBI-365 (a synthetic ABA analogue) reduced leaf crisping (section 2.2.4 for details in leaf crisping), which was due to increased sucrose accumulation on foliage, by reducing water loss (Pompodakis and Joyce, 2003). ABA, as spray, pulse and vase solution treatments, also extended foliage life of Geraldton waxflower by reducing solution usage and maintaining fresh weight (Joyce *et al.*, 1996).

ABA accumulates more in chloroplasts due to the high pH (Cowan *et al.*, 1982; Taiz and Zeiger, 1998). When ABA arrives at guard cells, it influences ion transport processes (i.e. efflux) that reduce guard cell turgor, thereby allowing the stomata to close and the plant to better retain water (Hartung *et al.*, 1998). ABA acts in the free space (apoplast) on the outer surface of guard cell plasma membranes (Pompodakis *et al.*, 2004). It inhibits an ATP-dependent proton pump activity in the plasma membrane of guard cells (Salisbury and Ross, 1992). This pump normally accelerates K^+ influx, which promotes stomata opening (Salisbury and Ross, 1992). Thus, ABA reduces K^+ transport, thereby causing stomatal closure in association with loss of guard cell turgor.

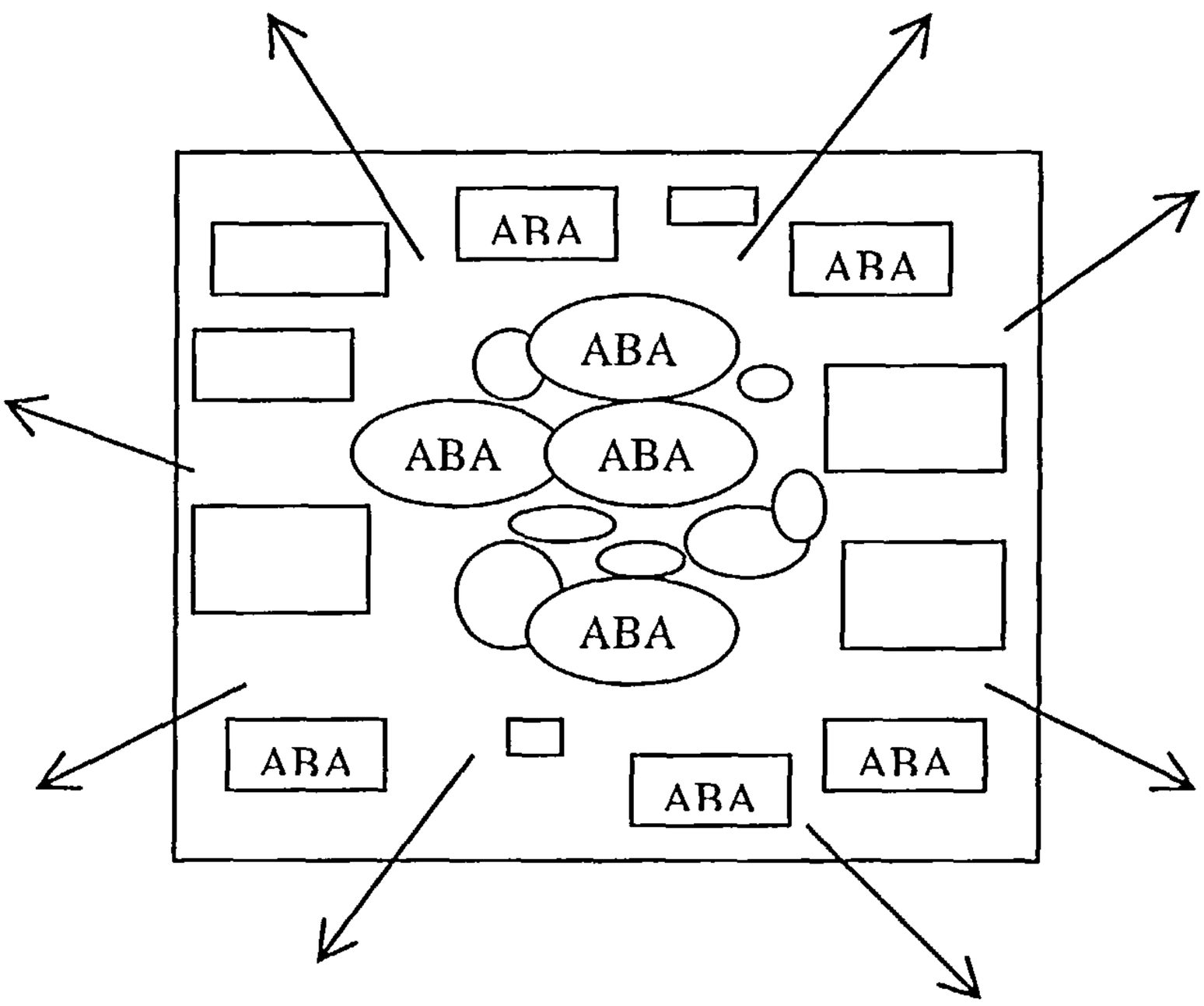
2.4.4 ABA distribution and its effects under water stress conditions

During the early stages of water stress, the pH of the xylem sap increases (Wilkinson and Davies, 1997). This increase influences the distribution of ABA between sub-cellular compartments in the leaf (Figure 2.3). An increase in xylem sap pH from 5.0 to 8.0 can enhance stomatal closure by changing ionisation state and,

thereby, the distribution of ABA (Wilkinson *et al.*, 1998). At lower pH, ABA is dissociated and tends to move from the apoplast into the symplast. At higher pH, ABA is not dissociated and remains in the apoplast where it can bind to ABA-receptors on the other face of the plasma membrane (Wilkinson *et al.*, 1998). Thus, the relative amount of endogenous ABA available outside the guard cells increases in plants under water deficit stress. This relative increase contributes to stomatal closure (Cowan *et al.*, 1982).

In cut roses, ABA levels increased in young isolated petals following water stress. This increase was attributed to ABA synthesis in petals (Le Page-Degivry *et al.*, 1991). When the ABA content in petals of daylily (*Hemerocallis* hybrid) flowers was increased under water stress, senescence-associated parameters, such as ion leakage, lipid peroxidation and proteinases and nucleases activities, were increased prematurely (Panavas *et al.*, 1998; Rubinstein, 2000). Recently, ABA was found to be involved in sucrose synthesis pathway in adult maize leaves under water stress conditions. Trouverie *et al.* (2003) reported that, ABA increased leaf vacuolar invertase activity; an enzyme involved in sucrose synthesis pathway. Among the four invertase genes studied (*Incw1*, *2*, *Ivr1*, *2*), only *Ivr2* gene expression was enhanced by ABA supply (Trouverie *et al.*, 2003).

A. pH 6 – ABA stored in symplast.



B. pH 7 – ABA remains in apoplast.

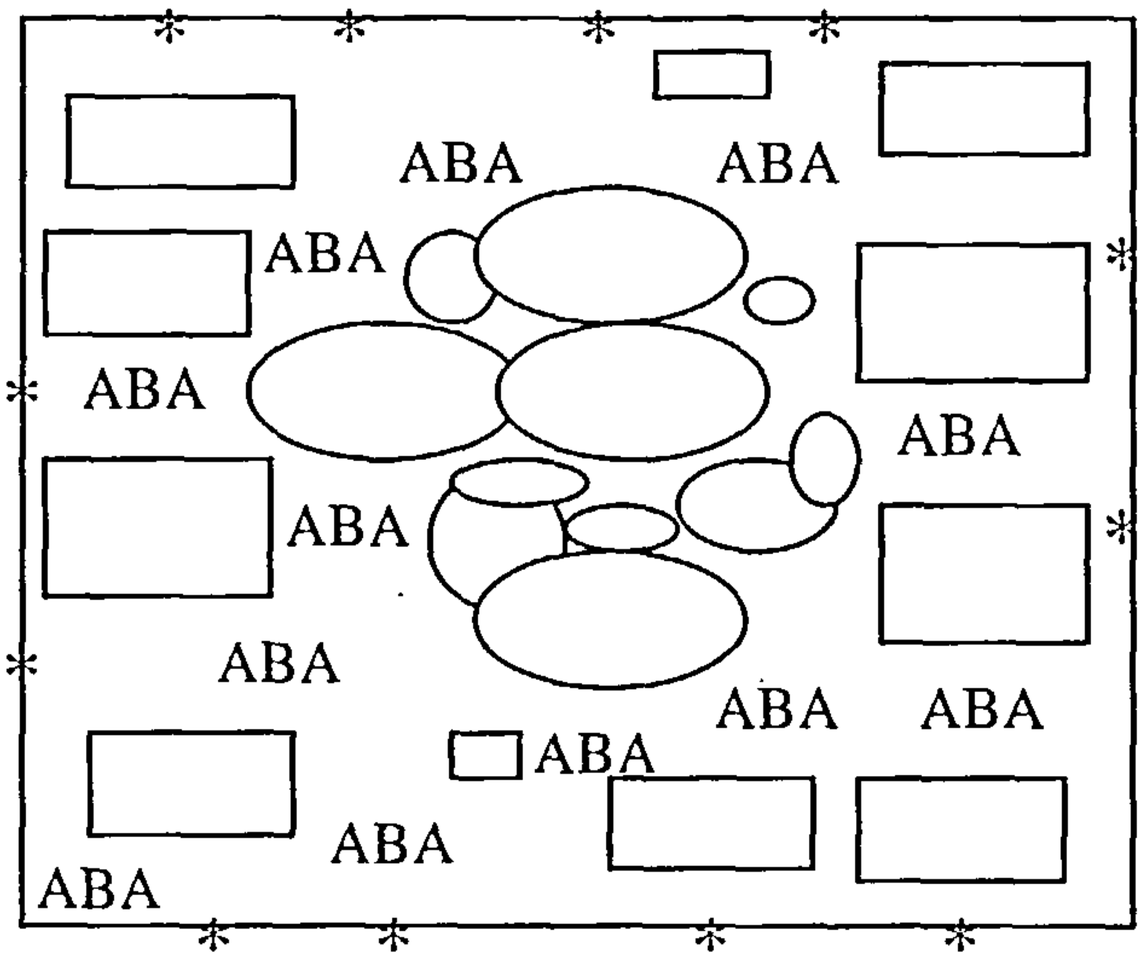


Figure 2.3: A schematic representation of the cells of a leaf from well-watered plant (A) removing ABA from the apoplast into the symplast. In contrast, in the cells of drought plants (B), ABA remains in the apoplast, where it can bind to ABA-receptors on the other face of the plasma membrane and cause stomatal closure (after Wilkinson, 1999).

2.5 CONCLUDING REMARKS

Cut flowers suffer from severe CI problems during storage leading to substantial economic losses (Come, 1991). Although roses are not highly sensitive to CI, storage at low temperatures causes irreversible symptoms (Low Tempeature Injury; LTI), such as incomplete flower opening and flower blueing, which are visible only after putting roses into vases (room temperature). Environment conditions during rose cultivation can substantially affect chilling tolerance. Depending on the cut flower species, extreme environment conditions, for instance, high (<25°C) or low (>15°C) temperature during cultivation, decrease vase life and quality parameters (Torre *et al.*, 2001). Flowers grown under these conditions and then stored at low temperature are possibly most sensitive to CI.

Research on water relations of cut flowers has indicated that water stress, which is due to continuous water loss (e.g. increased transpiration and/or stem blockage), leads to the visible end of vase life by enhancing senescence-associated changes. Today, solution treatments have been applied to cut flowers using different methods (e.g. pulse, vase solution and spay) for a range of purposes (e.g. carbohydrate supply, enhancement of water relation, microbial control). The plant growth regulator ABA promotes senescence in leafless flower stems, but, on the hand, it can induce stomatal closure protecting cut roses from water deficit stress (Pompodakis and Joyce, 2003; Pompodakis *et al.*, 2004). Endogenous ABA contents have been found to increase in plant cells under stress conditions, including chilling (Janowiak *et al.*, 2002).

CHAPTER 3

MATERIALS AND METHODS

3.1 GENERAL MATERIALS AND METHODS

3.1.1 Plant materials

Plants of rose 'First Red' (red petals) and 'Akito' (white petals) were grown in two different greenhouses on the island of Crete (Greece), between the latitudes 35°N and 25°E, for about two years (from April 2002 until June 2004). The greenhouses were found at both the north (Heraklion) GH1, and the south-eastern side (Ierapetra) GH2 of the island (Plate 3.1). Flowers were grown on beds (one bed per cultivar) using a hydroponic system. Rose plants were marked randomly in each bed and one flower stem of *ca.* 0.6 m was harvested from each plant at the tight bud stage. In each postharvest experiment, roses originated both from GH1 and GH2. In both glasshouses, roses were fertilised each day using liquid fertilisers at normal concentrations throughout the growth period (Appendix 3, Table A3.1). Electrical conductivity (E.C.) was greatest (3.5-4.0) in north greenhouse throughout growing period. Increase in E.C. was due to the poor quality of water in this region of Crete (Fthenaki, pers. comm., 2004). Irrigation volume varied from 2.5 (winter) to 3.5 (summer) l day^{-1} per m^2 of glasshouse surface area during the year

3.1.1.1 Flower preparation for storage

After harvest, flowers were stood in tap water and transported by car to the laboratory. The time period between harvest of flower stems and beginning of experiment was about 4 hours. Flower stems with 0.6 m length were sprayed with Rovral fungicide (1g AI iprodione l^{-1}) (Joyce and Shorter, 2000) and then left to air dry for about 10 min at the ambient laboratory temperature of 20°C. In order to determine the effect of storage temperature on subsequent vase life at 20°C, flowers were re-cut to a length of 0.5 m and placed in plastic boxes containing distilled water along with 10 mg l^{-1} dichloroisocyanuric acid (DICA) for wet storage. DICA was

used as antibacterial compound in order to prevent bacterial growth in the vase (van Doorn *et al.*, 1990; Knee, 2000). The rose flowers were then randomly put in temperature controlled cold-rooms.

3.1.1.2 Flower preparation for vase life experiments

Flowers of 0.4 m stem length bearing three leaves were used in all vase experiments. Two-cm was cut under water from the base of each stem to avoid air embolism (van Doorn, 1997) and flower stems were put into vases (day 0 of vase life). Flowers were stood in individual bottles containing 300 ml of vase solution. Vase solutions always contained 10 mg l^{-1} DICA. Bottles were washed out with distilled water at the end of each experiment and then left to dry. The top of each bottle was covered with cling-film held in place by an elastic band; thus, water loss could only occur via the flower (Mayak *et al.*, 1974). A slit was cut in the cling film to allow flowers to be inserted into vases. Vases were put randomly in the vase life room at the Technological Educational Institute (TEI) of Crete (Heraklion, Greece).

3.1.2 Glasshouse environment conditions

Air temperature (°C) and relative humidity (RH; %) data were collected for correlation with vase life parameters. Temperature and air humidity changes inside the glasshouses were measured continuously with tiny data loggers (Gemini Data Loggers, UK) during the year and data were downloaded using a GLM (Version 2.8) software. Two loggers (temperature and RH logger) were placed within the canopy of each glasshouse. The loggers were adjusted to record temperature and air humidity of the ambient environment every two hours during the year. Regional Photon Flux Density (PFD) data were collected every hour by the meteorological station of Technological Educational Institute (T.E.I) of Crete and correlated with vase life parameters.

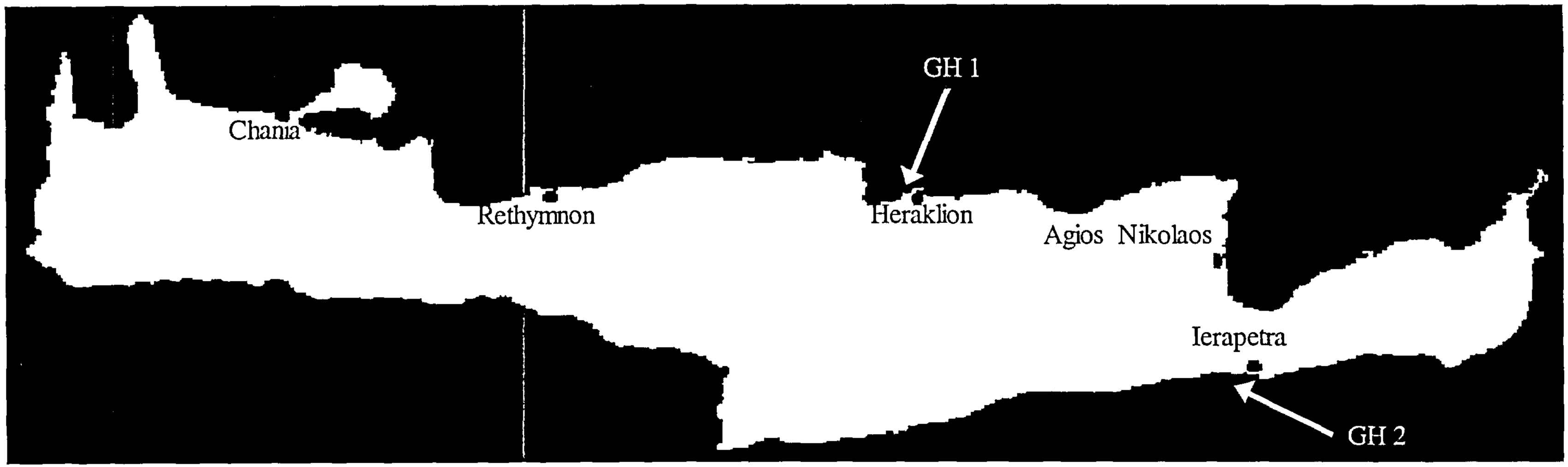


Plate 3.1: Location of Glasshouse 1 (GH1) and Glasshouse 2 (GH2) in the island of Crete.

3.1.3 Environment conditions in the cold- and vase life rooms

Rose flowers were stored in research cold-rooms in dark at 1 ± 0.5°C, 5 ± 0.5°C, and 10 ± 0.5°C. RH in the cold-rooms varied from 75% to 95% during storage experiments. Both temperature and RH were recorded daily using tiny data loggers. Vase experiments were carried out in the vase life room of T.E.I at 20 ± 5°C and 60 ± 10% relative humidity (RH). Temperatures and RH during each experiment were recorded using a calibrated thermo-hydrograph. The room was illuminated at 25 $\mu mol^{-2}\ s^{-1}$ at flower level using Sylvania 'cool white' fluorescent lamps on a 12 hours on-off cycle.

3.1.4 Assessments

Assessments were constantly performed before storage, after storage (d 0 of vase life) and during vase life in all experiments. Measurements were objective using the appropriate equipment (e.g. weighing balance, micrometer, chlorophyll fluorometer, colour meter) or subjective using rating scales and/or criteria that have been published in literature.

3.1.4.1 Vase life

Vase longevity was recorded as days of vase life from the time flowers were placed into bottles (day 0). Flowers were considered to terminate their vase life when the whole flower wilted or when advanced signs of fading were visible for most petals (Mayak and Halevy, 1974). Vase life (days) of leaves was judged to be over when > 50% on a stem had shrivelled. Chlorotic and necrotic areas were also considered to be the termination of foliage life.

3.1.4.2 Fresh weight changes and solution usage

Stem fresh weight (FW) and bottle weight (container + solution) were recorded at the same time every second day from d 0 of vase life. Data was used to calculate relative fresh weights (RFW) as percent of initial fresh weight (% RFW) and

solution usage as millilitres per gram initial fresh weight per second day (ml g^{-1} initial FW 2^{nd} d^{-1}), respectively (Joyce and Jones, 1992).

3.1.4.3 Corolla diameter

The maximum diameter of the flowers was measured every fourth day using a micrometer (MITUTOYO, Japan).

3.1.4.4 Bent neck incidence

When the peduncle of flower head was bent $\geq 90^{o}$ the symptom was recorded as bent neck. Visible observation of flowers, that were bent, was made daily. Data was used to calculate bent neck incidence (%) as percent of total replicate flowers for each treatment.

3.1.4.5 Chlorophyll fluorescence

Chlorophyll fluorescence parameters (F_0, F_m, F_v/F_m) were evaluated using a Handy Plant Efficiency Analyser (PEA) chlorophyll fluorometer (Hansatech instruments Ltd., King's Lynn, UK). Each replicate flower was measured after 15-20 min darkness at 20°C before and after cold storage and every 4^{th} day from the beginning of vase life. On removal from cold storage, flower stems were brought to the vase life room and equilibrated to 20°C for 15-20 min darkness before chlorophyll fluorescence values were re-measured. Chlorophyll *a* fluorescence is largely emitted by PSII at room temperature of about 20°C (Krause and Weis, 1984). At this temperature, approximately 95% of the chlorophyll fluorescence signal is derived from chlorophyll molecules associated with PSII. Dark adaptation of samples was achieved by covering the sample to be analysed, with a small, lightweight (approx. 6.2 gr), 7 mm diameter leafclip constructed from white plastic. The clip was closed using a small shutter blade so that light was excluded allowing dark-adaptation to take place (Plate 3.2).

Chlorophyll fluorometer calibration for roses

The period of darkness was experimentally determined using leaves of 'First Red' and 'Akito' roses. Fifteen clips were placed on leaves of each rose cultivar and the shutter plates were closed. All measurements were made on the upper side of the second five-leaflet under the flower head. Three measurements were carried out on each clip location in sequence at different time intervals between 1 and 30 minutes. Each clip was measured at 2-minute intervals using 100% light level. Chlorophyll fluorescence ratio (F_v/F_m) reading reached a peak at 16 and 18 minutes for 'First Red' and 'Akito' roses, respectively (Appendix 3, Table A3.2). Therefore, 20 minutes was deemed to be appropriate dark adaptation period for these cultivars. Having determined the dark adaptation period, the next stage was to determine the amount of actinic light required to fully saturate 'First Red' and 'Akito' roses. Six clips were placed on leaves of both cultivars with the shutter closed for at least 20 minutes dark adaptation period. Three measurements were made at each clip at increasing light intensities between 0 and 3000 μmol m^{-2} s^{-1}. The maximum F_v/F_m measurement was achieved at a light level of 2000 μmol m^{-2} s^{-1} (Appendix 3, Table A3.3).

3.1.5 Set up of experiments

Three different experimental sections are presented in the following chapters. The hypothesis that environmental changes can affect postharvest characteristics was investigated in the experimental section of chapter 4. Two genotypes of roses ('First Red' and 'Akito') were grown throughout the year (four distinct seasons; spring, summer, autumn and winter) and then their vase life parameters were evaluated. These roses were also stored at low temperature (1 and 5°C) for 10 days to examine the interactive effects of environmental changes and storage on vase life. In the experimental section of chapter 5, the same genotypes were grown from autumn to winter. Roses grown this period of year were most sensitive to CI. Thus, different ABA treatments (pulse, spray, control) were used before storage treatments (at 1°C and 5°C for 10 days, non-stored control roses) with the hypothesis of reducing CI and, thereby, promoting vase life. In the last experimental section (Chapter 5), 'Akito' roses, that were most sensitive to CI, were grown from the end of winter until the end

of spring. These roses were then stored at 1ºC for 10 days and novel ABA solutions (ABA, PBI-365, control) were applied to prevent CI.

Plate 3.2: Dark adaptation of rose leaves using leafclip before chlorophyll fluorescence measurement.

3.2 EFFECT OF SEASONAL VARIATION AND STORAGE ON VASE LIFE PARAMETERS OF 'FIRST RED' AND 'AKITO' ROSES

3.2.1 Plant materials

Flower stems of 'First Red' and 'Akito' roses were harvested from two different greenhouses (Chapter 3, Section 3.1.1). Eight different harvests were carried out during four seasons of the year 2002-03. Seasons were defined to be: autumn 2002 (September-November), winter 2002-03 (December-February), spring 2003 (March-May) and summer 2003 (June-August). One harvest was performed at the beginning and at the middle of each season, respectively, on the following dates: 1st of September and 15th of October (autumn 2002), 1st of December and 15th of January (winter 2002-03), 1st of March and 15th of April (spring 2003), 1st of June and 15th of July (summer 2003).

3.2.2 Experiment design

Eight vase life experiments were carried out during the year (2 experiments per season). In each experiment, harvested stems were stored wet at 1, 5, and 10°C for 10 days (Chapter 3, Section 3.1.1.1 for details in storage process). Ten replicate flowers of each variety were put in the vase life room directly after harvest as control treatment. Thus, eight factorial experiments consisted of two factors each were conducted. The examined factors of each experiment were: varieties ('First Red' and 'Akito' roses) and storage treatments (control, 1, 5 and 10°C). A Completely Randomised Design (CRD) was adopted in the vase life room and 10 replicate flowers were used for each treatment. A total of 80 flowers was used in each vase life experiment (160 per season).

3.2.3 Assessments

Flower and foliage condition was subjectively recorded during vase life using optical rating scales. A rating scale was also used to determine solution turbidity at the end of vase life. Furthermore, solution turbidity was objectively measured using a

spectrophotometer. Objective measurements were also performed to determine water loss and dry weight of detached leaves, petal colour and solution pH.

3.2.3.1 Flower condition

Flower development was assessed every 4th d by visual observation. The following flower development stages were defined: 1: Mature – tight bud, sepals partially released, 2: Early opening – about two whorls of petals released, the centre of flower still closed, 3: Mid opening – flower started to open, petals released, the centre of flower still closed, 4: Opened – open flower, stamens just visible, 5: Advanced opening – fully open flower, all stamens visible, petals with the first signs of colour fading and wilting (Plate 3.3).

Petal colour

Colour differences between flowers were recorded every 4th d of vase life using a colour meter Minolta GR-200. Three petals (external, middle and internal) were measured on each corolla. These petals were marked at the onset of vase life in order to be similar in each measurement. The CIELAB (L*, a*, b*) scale was used (McGuire, 1992). L*, a* and b* values pinpoint the measured colour in a three-dimensional colour space (Appendix 3, Plate A3.1). In the CIE (Commission Internationale de l' Eclairage; International Commission on Illumination) colour space, the lightness coefficient, L*, ranges from black = 0 to white = 100. On the horizontal axis, positive and negative a* indicates a hue of red-purple and bluish-green, respectively. On the vertical axis, positive b* indicates yellow and negative b* blue.

3.2.3.2 Foliage condition

The following leaf deterioration stages were defined: 1: Good turgor – leaves upright with firm turgor, 2: Softening – some loss of leaf turgor, 3: Slight wilting – all leaves showing slight wilting with softening, 4: Wilting: leaves lost turgor, bending and folding, 5: Senescence – leaves shrivelled, senescence of foliage. These stages were recorded every 4th d and plotted against time (Mayak and Halevy, 1974).

Plate 3.3: Five successive stages of rose flower opening: 1: Mature, 2: Early opening, 3: Mid opening, 4: Opened, 5: Advanced opening.

3.2.3.3 Water loss and dry weight of detached leaves

Water loss of rose leaves was determined after detachment at 75, 85 and 95% RH and 1, 5 and 10°C. In order to achieve three different RH levels, sodium chloride, potassium chloride and potassium nitrate, that give 75, 85 and 95% RH, respectively, were diluted individually in distilled water until saturation point (Greenspan, 1997). Afterwards, the super saturated salt solutions were placed in boxes of 10 L. Salt saturated solutions produce the desired RH of a closed space, by changing vapour pressure (Greenspan, 1997). RH was measured in the free space of boxes using a RH data logger to confirm that RH levels were correct.

At the time of harvest, flower stems were wrapped in newspapers moistened with distilled water and then over-wrapped with a plastic tissue. The wrapped stems were transported by car to the laboratory four hours after harvest and unwrapped at the ambient temperature of 20°C. One five-leaflet leaf was detached from the middle of rose stem. The leaves were then placed in the free space of boxes, which contained the saturated salt solutions. The boxes were covered with vaseline and then over-covered with parafilm to avoid evaporation. These leaves were weighed 0, 12, 24, and 48 hours after detachment and the water loss was calculated as percent of total water content (water loss %) (Mortensen and Fjeld, 1995; Mortensen and Gislerod, 1999). Leaves were then dried at 60°C for 48 hours for determination of dry weight (Mortensen and Fjeld, 1995). At each harvest, five replicate leaves were used from each cultivar for each treatment. Eighteen different treatments (3 temperatures x 3 RH levels x 2 cultivars) were used in each experiment. Thus, a total of 180 leaves (90 leaves per experiment x 2 replicate experiments) was used each season.

3.2.3.4 Solution pH

Solution pH was measured with a pH meter (Crison instruments Ltd., 1990) at the beginning and at the end of each experiment. The meter was calibrated at 20°C with pH 7 and 4 ± 0.01 buffer solutions (Russell pH Ltd., UK). After calibration, solution pH was measured in each replicate vase.

3.2.3.5 Solution turbidity

Solution turbidity was determined subjectively at the end of each experiment. The following stages of solution cloudiness were defined: 1: Clear, 2: Slightly cloudy, 3: Cloudy (Plate 3.4) (Pompodakis *et al*., 2004). Solution turbidity was also measured at the end of each experiment at the optical density (OD) of 400 nm, 500 nm, and 600 nm with a spectrophotometer (Hitachi V-200) calculating the mean of these three values (Knee, 2000). Distilled water was used as a blank.

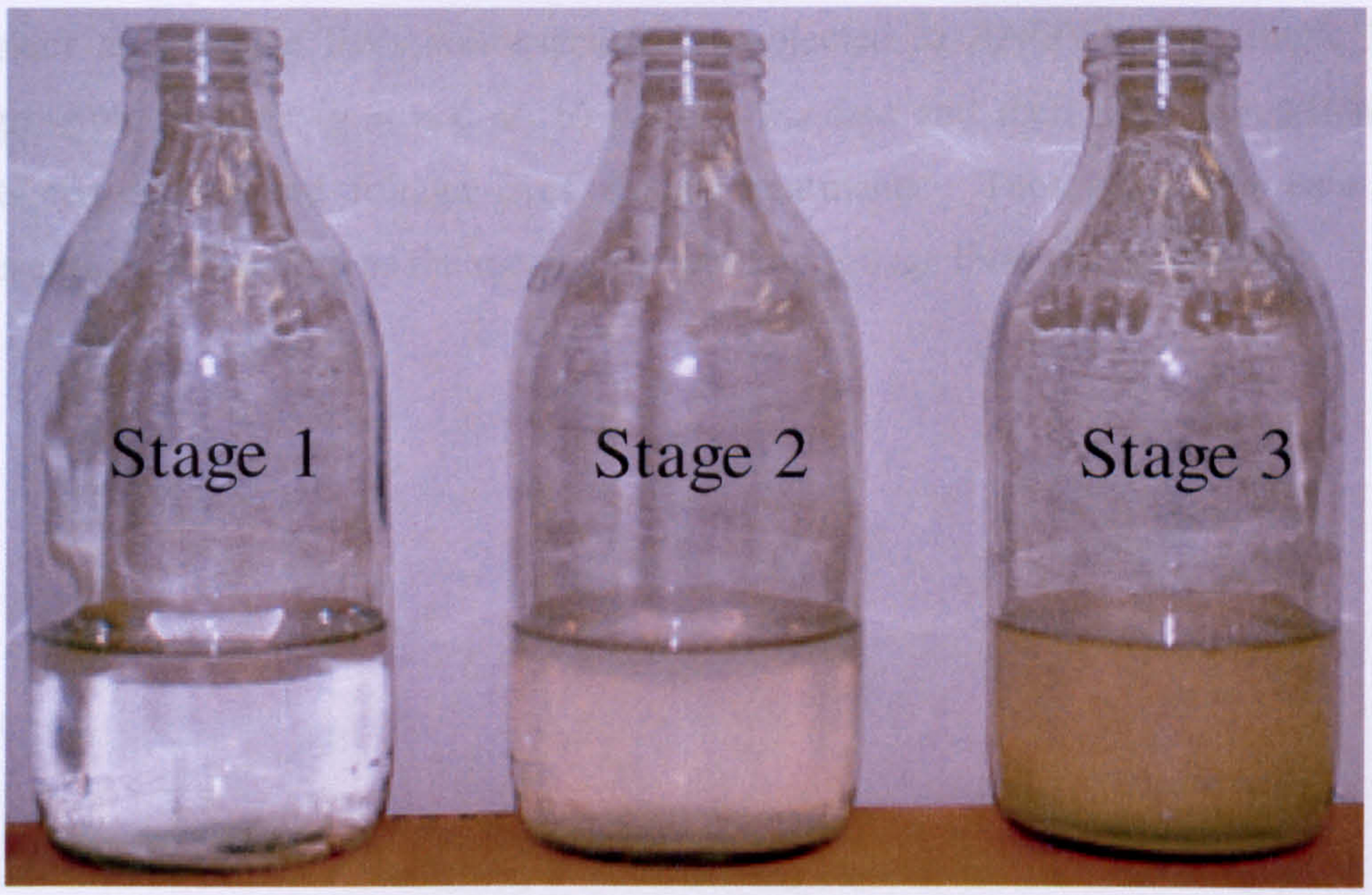


Plate 3.4: Three successive stages of vase solution turbidity: Stage 1 = clear, Stage 2 = slightly cloudy, and, Stage 3 = cloudy (after Pompodakis *et al*., 2004).

3.2.4 Statistical analysis

Data were processed by Analysis of Variance (ANOVA) model (univariate ANOVA) to compare main factor means. One-factor data were also analysed by one-way ANOVA. Individual treatments means were compared using the Duncan's multiple range test at P = 0.05 (Field, 2000). The non-parametric Kruskal-Wallis (3 or more levels) test was used to determine differences in flower and foliage stages

within factors (Little, 1985). In the cases that non-parametric tests were similar to ANOVA for non-parametric data (e.g. turbidity rating scoring), only ANOVA tables were presented in appendices. Correlations between vase life duration and vase life parameters (i.e. F_v/F_m, flower stage, foliage stage, fresh weight and solution usage) and between growing conditions (i.e. temperature, RH and PFD) and vase life variables (i.e. vase life, F_v/F_m, flower stage and solution turbidity) were carried out using Pearson's correlation test. Statistical analysis was performed using SPSS 9.0 (Statistical Package for the Social Science, Chicago, IL, USA) for Windows. Linear regression analysis and graphic presentation was performed in Sigmaplot 2000 (Chicago, IL, USA) for Windows. Data in text are presented as main factor means in tables and the corresponding individual treatment means are presented in figures. The results of the statistical tests are presented in appendices.

Flower and foliage lives were separately subjected to ANOVA and simple linear regression analysis (y = a.x +/- b) with F_v/F_m data and there were no differences between flower and foliage lives among treatments. Thus, vase life data were presented in Chapter 4 as the mean of flower and foliage lives (Knee, 2000).

3.3 EFFECT OF DIFFERENT ABSCISIC ACID TREATMENTS TO IMPROVE VASE LIFE OF CUT 'FIRST RED' AND 'AKITO' ROSES STORED AT LOW TEMPERATURE

3.3.1 Plant materials

Rose 'First Red' and 'Akito' stems grown to the tight bud stage in GH1 and GH2 were used in these experiments (Chapter 3, Section 3.1.1 for details in growing conditions). Four different harvests were performed from the beginning of autumn 2003 (September-November) until the end of winter 2003-04 (December-February). One harvest was performed at the beginning and at the middle of each season, respectively, on the following dates: 1st of September and 15th of October (autumn 2003), 1st of December and 15th of January (winter 2003-04). These two seasons were selected according to the results of chapter 5. Roses grown from autumn to winter appeared to be most sensitive to low temperature storage.

3.3.2 Experiment design

Four postharvest experiments (two during autumn and two during winter) were carried out to determine the interactive effects of different ABA treatments and low temperature storage on vase life of 'First Red' and 'Akito' roses grown from autumn to winter. In the 1st autumn experiment, 'First Red' roses received three different ABA treatments after harvest. One group of flower stems was sprayed on the foliage with 10^{-5} M ABA at 20°C, another group was pulsed with 10^{-1} M ABA for 24 h at 20°C under a light intensity of 25 μmol^{-2} s^{-1} at flower level on a 12 hours on-off cycle and a last group was used as control (without ABA treatment). After receiving the three ABA treatments, roses of each group were stored wet at either 1 or 5°C for 10 days or put in the storage rooms directly after harvest as control treatment. Thus, a factorial experiment, consisting of three different treatments before storage (pulse, spray and control) plus nine treatments after storage (three ABA treatments x three storage treatments), was conducted after harvest.

In each treatment, rose stems were used to conduct non-destructive and destructive measurements on d 0 and d 10 of vase life. Three replicate roses from

each treatment were used to conduct non-destructive measurements throughout vase life (e.g. fresh weight, vase solution weight, vase life, corolla diameter, and chlorophyll fluorescence). Six replicate roses were also used to conduct the destructive essays of electrolyte leakage and lipid peroxidation (three roses for d 0 and three roses for d 10 of vase life). Thus, nine replicate roses were used for each treatment to conduct the non-destructive and destructive measurements. A total of eighty-one roses (nine replications x nine treatments) was used and a CRD was adopted in the vase life room.

The same experiment was repeated during the autumn (section 3.3.1 for details in 2nd autumn harvest) using 'Akito' roses instead of 'First Red'. This experimental structure was also followed during the winter experiments in order to determine effects of growing season. Additionally, in the last winter experiment with 'Akito' roses, six replicate roses (three roses for d 0 and three roses for d 10 of vase life) were used for each treatment to conduct the ABA essays (destructive measurement).

3.3.3 Solution preparation

ABA was dissolved in a minimal amount of ethanol (*ca.* 1 ml) and then diluted in distilled water to the final working concentration of 10^{-5} M and 10^{-1} M for spray and pulse treatments, respectively. The final concentration of ethanol in both solutions was 0.1% (v/v). Solutions during wet-storage and vase solutions also contained 10 mg l^{-1} DICA (Chapter 3, section 3.1.1.1 for details in storage process and section 3.1.1.2 for details in vase life experiments).

3.3.4 Biochemical assays

3.3.4.1 Membrane leakage

Membrane leakage was determined by measurement of electrolyte leaked from leaves and petals. Electrolyte leakage (EL) was measured according to the method of Karabal et al. (2003) with slight modifications. Briefly, two five-leaflet leaves (2nd and 3rd leaves from the top of the stem) and 2-3 petals (selected randomly from corolla) were detached from each flower stem and weighted immediately. Three replicate stems were used from each treatment. Each sample (leaves or petals) of *ca.*

3 g F.W. was placed in a 50 ml conical flask containing 5 ml of 0.4 M mannitol solution and shaken gently on an automated shaker for 3 h at 20°C. After incubation, the electrical conductance of solution was measured (C_1) using a conductance meter (Crison instruments Ltd.). The samples were then autoclaved for 10 min at 120°C to inactive the tissue completely and the second conductance (C_2) was measured to determine the ion concentration after complete membrane disintegration. The total injury to the membranes was expressed as the percentage of the initial conductivity over the total conductivity according to the formula (C_1/C_2) x 100.

3.3.4.2 Lipid peroxidation

Lipid peroxidation in rose leaves and petals was determined by estimating the malondialdehyde (MDA) content using the thiobarbituric acid (TBA) reaction as reported by Piqueras *et al.* (2002) for carnation leaves with slight modifications. Samples of 1 g F.W. of leaf (4^{th} and/or 5^{th} leaf from the top of the stem) and petal (selected randomly from corolla) tissue were homogenised in 5 ml of 0.1% trichloroacetic acid (TCA) (w/v) with an Ultra Turrax T 25 S7 homogeniser (Janke & Kunkel GMBH & Co. KG IKA Labotechnik) at 8000 rpm for 1 min. The homogenate was centrifuged (Hellenic Labware Centrifuge, K – 80, GR) at 10,000 rpm for 5 min and 4 ml of 20% TCA containing 0.5% TBA (w/v) were added for every 1 ml of the aliquot of the supernatant. The mixture was heated at 95°C for 30 min on water bath and then cooled quickly on ice bath. The resulting mixture was centrifuged at 10,000 rpm for 15 min and the absorbance of the supernatant was measured at 532 nm and corrected for non-specific turbidity by subtracting the absorbance at 600 nm. 0.5% TBA in 20% TCA solution was used as the blank. MDA content was determined using the extinction coefficient of 155 mM^{-1} cm^{-1} (Heath and Parker, 1968).

3.3.4.3 ABA determination

Sample preparation for ABA determination

Flower petals and the two uppermost five-leaflet leaves were detached from the same flower stem. Samples for ABA determination were collected before storage,

on d 0 and d 10 of vase life (section 3.3.2 for details in experimental design). These rose materials were snap frozen in liquid nitrogen, lyophilised and freeze-dried at -50°C for 24 hours using a freeze-drier (Heto-Holten A/S., FD 8.0, Denmark). The freeze-drier was pre-cooled to -20°C to avoid thawing of samples. Freeze-dried plant tissues were then stored at -50°C for two months until ABA extraction.

Extraction procedure

All procedures for ABA extraction and purification were carried out under conditions of low light intensity and temperature to minimise photodegradation and oxidation of the phytohormone (Montero *et al.*, 1994). ABA was extracted as described by Carrasquer *et al.* (1990) for crude plant extracts with some modifications. About 500 mg freeze-dried rose tissues were shaken in 150 ml of methanol of HPLC grade-6.8 mM phosphoric acid (80:20, v/v) containing 100 mg l^{-1} of 2,6-di-t-butyl-4-methyl phenol (BHT) as an antioxidant at 4°C in the dark for 24 h on a shaker. The homogenate was filtered through Whatman No. 3 filter paper in a 5.5 cm in diameter Bucher Funnel under vacuum filtration. The remaining fraction was again extracted with 100 ml of methanol-6.8 mM phosphoric acid (80:20, v/v) for 4 h under the same conditions (at 4°C in the dark for 24 h) until the plant material became colourless. The filtrate was adjusted at pH 8.5 with 0.6 M sodium hydrogen carbonate and reduced in a Buchi rotary evaporator at 35°C to an aqueous phase. The aqueous phase was frozen at -20°C until the pre-purification procedure.

Pre-purification procedure

In the pre-purification step, the aqueous phase was left to thaw at the ambient temperature of *ca.* 23°C for 5 min and then poured into a beaker. The pH was adjusted to 2.6 with concentrated phosphoric acid (8.6 M) and loaded with a syringe onto an RP-18 cartridge (40-63 μm, LiChrolut®, Merck), pre-wetted with 5 ml of methanol and then with 5 ml of water. The cartridge was washed with 5 ml of methanol-6.8 mM phosphoric acid (30:70, v/v) to elute the most polar compounds. The cartridge was then washed with 3 ml of methanol-6.8 mM phosphoric acid (60:40, v/v) and the eluted fraction was evaporated to dryness in a freeze-drier.

ABA purification and further quantification by HPLC

The dried sample was dissolved in 250 µl of methanol-6.8 mM phosphoric acid (50:50, v/v) and 200 µl were put in HPLC vials. Individual vials were loaded into a Kontron Instruments HPLC 360 autosampler, connected to a 335 dual UV detector, attached to a 352 pump (Sci-Tec Instruments, Beds, UK) for ABA separation and further quantification. The HPLC column was C_{18} (30 x 0.4 cm i.d., Waters Associated). Solvent A was water and solvent B methanol. Both solvents contained 1% acetic acid. Elution was conducted at a flow rate of 1.2 ml min^{-1} with a 30 min linear gradient starting with 20% B and ending with 80% (Wang *et al.*, 2002). The detector was set up at 260 nm.

Calibration graph

Standard ABA solutions (2-cis, 4-trans abscisic acid; pKa = 4.8; Sigma Chemical Company) of 0.1, 1.0, 10, and 100 ng μl^{-1} in methanol-6.8 mM phosphoric acid (1:1, v/v) were prepared and three replicates of each were injected into the HPLC to determine retention times for ABA (Appendix 3, Table A3.4). The mean retention time of ABA was 13.62 (± 0.26) min and the HPLC was adjusted to collect the biggest peak in this range of retention time. ABA content in rose leaves and petals was identified by comparison of samples with the retention time of standards. For the calibration graph, amounts from 0.1 to 1600 ng μl^{-1} were used, as this is the range of endogenous ABA levels that have been identified previously in tissue of roses (Le Page-Degivry *et al.*, 1991; Muller *et al.*, 1999). Standard solutions containing 0.1, 10, 100, 200, 400, 800 and 1600 ng μl^{-1} of ABA in methanol-6.8 mM phosphoric acid (1:1, v/v) were prepared and three replicates of each were injected into the HPLC system. ABA concentration was linearly correlated ($r^2 = 0.99$) with the peak areas (Figure 3.1). A calibration run was made every time samples were run.

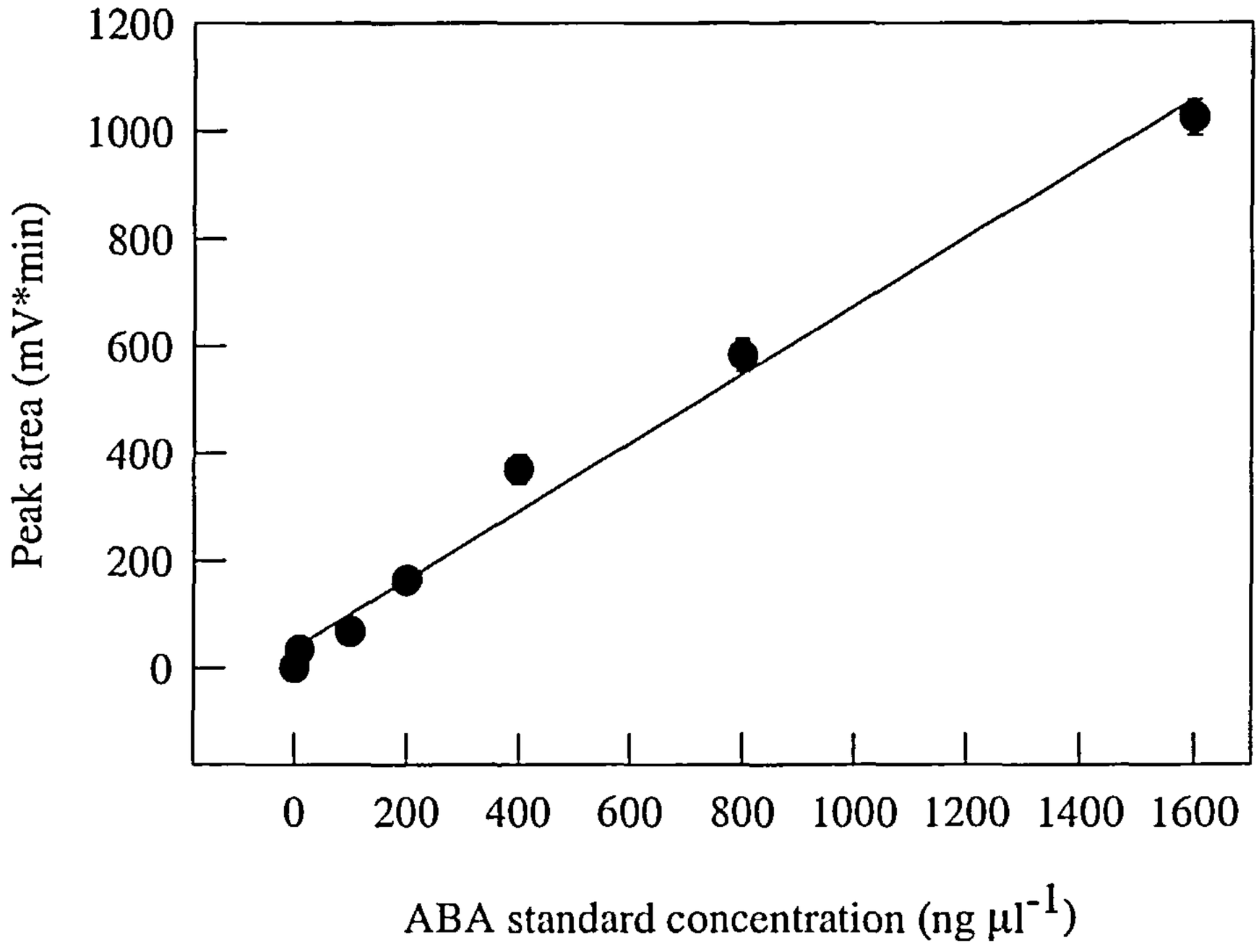


Figure 3.1: Calibration curve of ABA standard concentration (0.1, 10, 100, 200, 400, 800 and 1600 ng μl^{-1}) vs. peak area (mV*min). Data are means; n = 3. Regression line: $y = 0.63x + 36$; $r^2 = 0.99$. Bars indicate the SE (n = 3).

3.3.5 Light microscopy

3.3.5.1 Rose sample fixation

At the time of harvest (tight bud stage), three replicate peduncles of 5 cm-length were cut from stems of 'First Red and 'Akito' roses, respectively. The peduncles were fixed in formalin – acetic acid – ethanol (F.A.A.) solution made of 5 ml glacial acetic acid and 90 ml 70% ethanol (v/v) (Purvis *et al.*, 1966). The solution was made immediately before use to avoid chemical reaction between the acid and the alcohol (e.g. ethyl acetate production), which would have resulted in unstable fixative.

3.3.5.2 Sample dehydration and clearing

Fixed peduncles were dehydrated, cleared and infiltrated in the series of ethanol, toluene and wax immersions (Table 3.1). The fixed samples were dehydrated using a series of increasing concentration of ethanol to avoid shrinkage of the materials. After dehydration, the ethanol was replaced with toluene, which is miscible with alcohol, to clean the samples (Purvis *et al.*, 1966).

Table 3.1: Steps for rose peduncle dehydration and clearing.

Step	Solvent (v/v)	Time (h)
1	80% ethanol	3
2	90% ethanol	2
3	100% ethanol	2
4	100% ethanol	2
5	toluene	2
6	toluene	4
7	paraffin wax	2
8	paraffin wax	2

3.3.5.3 Embedding, making the block and sectioning

Cleared peduncles were embedded in a bath of molten wax at 50°C for 12 h. The wax gives the support to the tissues, which is necessary before sections can be cut in the microtome (Purvis *et al.*, 1966). After infiltration with wax, the peduncles were cast into a block of fresh wax, which was then stored at 3°C for 48 h until sectioning. Transverse sections of 10 μm thick were cut with a microtome carrying a steel knife (Bright Instrument Co. Ltd., Huntingdon, UK) from the same relative positions in 'First Red' and 'Akito' peduncles. The transverse sections were cut at one quarter, half and three quarters of the way along the peduncles (D.C. Joyce, Pers. Comm., 2004) and, thus, three transverse sections were taken from each replicate peduncle.

After cutting, the sections were floated-out on warm water (*ca.* 50°C), in which gelatin (0.1% v/v) had been dissolved to act as an adhesive, and left to dry at 30°C for 12 h.

3.3.5.4 Section staining

Before staining, sections were first de-waxed in toluene for 10 min, dehydrated in 50% and 70% (v/v) ethanol for 2-5 min per step and washed with distilled water. Sections were then stained with phloroglucinol-HCl, which stains lignified cell walls red-orange (Zhong *et al.*, 2000). They were dipped in phloroglucinol for 10 min and then a drop of concentrated HCl was added in each section. Finally, the sections were mounted in DPX and the samples were examined under a Leitz Laborlux K compound microscope fitted with cool light illuminator.

For examination of samples under fluorescent light, sections were stained with Safranin O for 24 h (Ruzin, 1999) and then washed in distilled water to remove excess stain. Sections were dehydrated in 50% and 70% (v/v) ethanol for 2-5 min per step. They were then counterstained in Fast Green for 10 sec, dipped rapidly through an ethanol (80, 90, 100 and 100% v/v) series to remove traces of water, cleared in toluene and finally mounted in DPX. Samples were examined under Ploempak incident light fluorescence illuminator with filter blocks I_2 (exciting filter 450-490, beam splitter 510, barrier filter 515 and exciting filter 340-380, Beam splitter 400, barrier filter 430) (Leica Microsystems, Milton Keynes, UK) using different stains.

3.3.5.5 Histological assessments

The diameter of each peduncle was measured using a micrometer (MITUTOYO, Japan) at the time of sectioning. The diameter of each vascular bundle was also measured under a Leitz Laborlux K compound microscope fitted with cool light illuminator and an eyepiece graticule, which had been calibrated using a haemocytometer slide for each combination of microscope lens and microscope objective. These data were used to calculate the proportion (%) of transverse section area that was vascular bundles (D.C. Joyce, Pers. Comm., 2004). The absolute numbers of vascular bundles were measured in each of the transverse section. The numbers of xylem elements was also counted in a random sample of 10 vascular

bundles to calculate number of xylem elements per vascular bundle in each transverse section.

3.3.6 Statistical analysis

Data were processed by Analysis of Variance (ANOVA) model (univariate ANOVA) to compare main factor means. A test probability level of $P < 0.05$ was used. One-factor data (F_v/F_m, corolla diameter, fresh weight, solution usage during vase life and histological parameters) were also analysed by one-way ANOVA. Individual treatments means were compared using the Duncan's multiple range test at $P = 0.05$ (Field, 2000). Correlations between vase life parameters (i.e. F_v/F_m, electrolyte leakage, MDA, solution usage on day 7, corolla diameter and fresh weight on day 8) and flower or foliage lives were carried out using Pearson's correlation test. ABA content in petals and leaves of 'Akito' roses was also correlated with flower and foliage lives, respectively. Statistical analysis was performed using SPSS 9.0 (Statistical Package for the Social Science, Chicago, IL, USA) for Windows, while graphic presentation was performed in Sigmaplot 2000 (Chicago, IL, USA) for Windows. Data in text are presented as main factor means in tables and the corresponding individual treatment means are presented in figures. The results of the statistical tests are presented in appendices.

3.4 EFFECT OF ABA AND ABA ANALOGUE TREATMENTS, BEFORE AND AFTER STORAGE AT 1°C, ON VASE LIFE OF CUT 'AKITO' ROSES

3.4.1 Plant materials

Rose cv. 'Akito' was grown to the tight bud stage in GH1 and GH2 (Chapter 3, Section 3.1.1 for details in growing conditions) from the end of winter 2003-04 (February) until the middle of spring 2004 (15th of April). One harvest was performed at the end of winter (February) and another at the middle of spring 2004 (April). These two periods of harvests were selected to be close to winter.

3.4.2 Experiment design

Two replicate postharvest experiments were conducted using the ABA analogue 8-methylene ABA methyl (PBI-365; Plant Biotechnology Institute, National Research Council of Canada, Saskatoon, S7N 0W9, Canada). Flower stems of 'Akito' roses, which are most sensitive to CI, received two different treatments after harvest. One group of flowers was pulsed with 10^{-1} M ABA for 24 h while another group was put in the storage rooms directly after harvest as control treatment. Flowers were then stored wet at 1°C for 10 days (section 3.1.1.1 for details in storage process). After storage, roses of each group were put into vases containing 3 different solutions; 10^{-5} M ABA, 10^{-5} M ABA analogue (PBI-365) and distilled water as control treatment. All vase solutions contained 10 mg l^{-1} DICA as antimicrobial compound (section 3.1.1.2 for details in vase life experiments). Thus, a factorial experiment consisting of two factors (two pulse solutions x three vase solutions) was arranged in a CRD in the vase life room.

Nine replicate roses (three roses for non-destructive measurements, three roses for EL and MDA, three roses for ABA essays) were used for each treatment to conduct non-destructive and destructive measurements, including the ABA assays (section 3.3.2 for details in the number of replications). A total of fifty-four (nine replications x six treatments) replicate flowers was used in this experiment. The same experimental structure was adopted in the second replicate experiment, in which ABA was not measured and, therefore, six replications were used for each treatment.

3.4.3 Solution preparation

Both ABA the analogue PBI-365 were dissolved in ethanol and then diluted in distilled water to the final working concentration. The final concentration of ethanol in both solutions was 0.1% (v/v). Solutions during wet-storage and vase solutions also contained 10 mg l^{-1} DICA (Chapter 3, section 3.1.1.1 for details in storage process and section 3.1.1.2 for details in vase life experiments).

3.4.4 Assessments

Assessments were performed as described in section 3.1.4. Biochemical assays of EL, MDA and ABA were performed as described in section 3.3.4. ABA assays were carried out in the first replicate experiment only.

3.4.5 Statistical analysis

One-factor data were analysed by one-way ANOVA. Individual treatments means were also compared using the Duncan's multiple range test at P = 0.05 (Field, 2000). In graphs, LSD (P = 0.05) tests were applied for mean separation. Statistical analysis was performed using SPSS 9.0 (Statistical Package for the Social Science, Chicago, IL, USA) for Windows. Linear regression analysis and graphic presentation was performed in Sigmaplot 2000 (Chicago, IL, USA) for Windows. The results of the statistical tests are presented in appendices.

CHAPTER 4

EFFECT OF SEASONAL VARIATION AND STORAGE TEMPERATURE ON VASE LIFE PARAMETERS OF 'FIRST RED' AND 'AKITO' ROSES

4.1 INTRODUCTION

4.1.1 Storage of cut roses grown in different seasons

The vase life of cut roses often depends on growing season (Urban *et al.*, 1995). Seasonal variation of pre-harvest environmental conditions, such as temperature, air humidity, light intensity and lighting period during cultivation, can influence the vase life of cut roses (Shin *et al.*, 2001). Vase life generally declines for roses grown in autumn and winter months (from September until February [Northern Hemisphere]) due to early flower wilting and leaf problems (e.g. leaf crisping; Chapter 2, section 2.2.4) (Slootweg *et al.*, 2001). Moreover, vase life parameters in roses, such as bent neck incidence, transpiration rates and flower opening, are seasonally dependant (Mortensen and Fjeld, 1995). However, the effect of seasonal variation on chilling tolerance of roses during storage has not been studied extensively.

Postharvest storage of roses at low temperature is a useful practice, in terms of regulating market flow (Nowak and Rudnicki, 1990). However, a reduction in vase life and loss of flower quality has been recorded after short- or long-term storage at 0-4°C (Faragher *et al.*, 1986; Serrano *et al.*, 1992). Many of the post-storage disorders observed in roses, such as incomplete flower opening (Faragher *et al.*, 1986), petal bueing (Leonard *et al.*, 2001) and foliage wilting (Hu *et al.*, 1998a), are partially related to chilling-induced alterations in metabolic processes. Primary events of chilling stress seem to include changes in membrane functions (re-distribution of membrane lipids and proteins) as a response to alterations in the membrane properties (Come, 1991; Marangoni *et al.*, 1996). The development of visible symptoms is a result of secondary processes that are enhanced by higher temperatures (van Kooten *et al.*, 1992).

4.1.2 Chlorophyll fluorescence and low temperature injury

Low temperature stress causing low temperature injury (LTI) in roses cannot be readily assessed by visual observation. Many instrumental techniques for assessing stress effects are destructive (Schapendonk *et al.*, 1992). Chlorophyll fluorescence (CF) has been used to study cellular processes, other than photosynthesis, the possible responses of plants to various stresses, including chilling tolerance in particular (Brennan and Jefferies, 1990; Walker *et al.*, 1990; Hakam *et al.*, 2000; Rosenqvist and van Kooten, 2002). Therefore, CF may have potential for rapid quantitative assessment of LTI in roses. CF is sensitive and non-destructive, and can detect injury before visible symptoms appear (van Kooten *et al.*, 1992; Hakam *et al.*, 2000).

CF from higher red to far-red wavelengths provides information on physicochemical condition of the photosynthetic apparatus (Krause and Weis, 1984). In dark-adapted plant tissues, the electron transport pathway of PSII is open because Q_A, the primary electron acceptor of PSII, is fully oxidised. As a result, fluorescence is minimal (F_0). If continuous illumination is commenced, CF temporarily achieves a maximum (F_m) due to Q_A reduction (Krause and Weis, 1991). The difference between maximum and minimum fluorescence ($F_m - F_0$) is termed variable fluorescence (F_v). The ratio F_v/F_m is used to provide a diagnostic measure of the overall photosynthetic efficiency with which green plant tissue is able to utilise light. A decline in F_v/F_m after a short dark adaptation period is an indication of photoinhibitory damage in plants subjected to chilling (Aroca *et al.*, 2001).

In fruits and vegetables, CF has been reported to have potential in measuring chilling stress. After 3 months of storage, 'McIntosh' apples (*Malus domestica* Borkh.) stored at 0°C had lower F_v values than similar apples stored at the optimum temperature of 3°C, indicating the development of LTI (DeEll *et al.*, 1995). CF was also a good predictor of LTI in green bell peppers before tissue damage became visible (Lurie *et al.*, 1994). F_m/F_0 of green peppers (*Capsicum annuum* L.) stored at 2°C decreased 9.0-fold during the first week when the surface pitting was not visible (Lurie *et al.*, 1994). Furthermore, van Kooten et al. (1992) found that F_v/F_m of cucumber fruit (*Cuccumis sativus* L.) stored for 2 weeks at 10°C did not change during storage, while cucumbers stored < 7°C exhibited a significant decrease in

F_v/F_m, along with discoloration and increased decay incidence. The decrease in F_v/F_m was temperature dependent and was even more pronounced after an additional 6 days at 20°C.

CF was found to be a suitable method for determining chilling tolerance in rose plants (Hakam *et al.*, 2000). Thus, this technique has potential for quality assessments in nurseries and breeding programs. However, detection of LTI in cut roses after cold-storage using CF has not been evaluated. F_v/F_m has been used to detect LTI after storage in harvested kangaroo paw (*Anigozanthos* sp.) inflorescences (Joyce and Shorter, 2000; Miranda *et al.*, 2000). Reductions in F_v/F_m after storage were linearly correlated with declining post-storage vase life (Miranda *et al.*, 2000).

This study was undertaken in an attempt to explain the variation of vase life parameters (e.g. CF, vase life duration, flower opening, fresh weight, solution usage and vase solution parameters) in roses grown from Autumn 2002 to Summer 2003 and then stored at low temperature (Chapter 3, sections 3.1.4 and 3.2). Seasonal changes (e.g. temperature and relative humidity) during cultivation were recorded in the rose production glasshouses in Crete while photon flux density (PFD) records were collected from the nearest weather station in Heraklion (Chapter 3, section 3.1.2). The aim of the present study was to investigate whether environmental changes and storage treatment could affect the overall quality of cut roses (e.g vase life parameters). In parallel with vase life parameters, CF was measured (Chapter 3, section 3.1.4.5) to determine whether it could provide a rapid and simple method for assessing LTI in cut roses. CF was also correlated with seasonal changes and vase life parameters.

4.2 RESULTS

4.2.1 Interactive effects of growing season and storage temperature on vase life

4.2.1.1 Vase life and F_v/F_m

Growing season, storage temperature and their interaction had significant effects ($P \leq 0.001$) on vase life duration (Table 4.1, Appendix 4.1.1, Table A4.1.1.1 and A4.5.1.5). As a main factor mean for all storage treatments, 'First Red' and 'Akito' roses grown in summer averaged greatest vase lives of 10.9 and 11.9 days, respectively. However, for both cultivars grown during winter, vase life decreased markedly to 6.0 days for 'First Red' and 7.4 days for 'Akito'. Vase lives of roses grown during spring and autumn were intermediate. Shorter vase lives during winter were recorded for both non-stored and stored flowers (Figure 4.1A, B). Storage of roses significantly ($P < 0.05$) reduced vase life compared to non-stored control flowers throughout the year (Table 4.1, Appendix 4.1.1, Tables A4.1.1.9 – A4.1.1.16). The reduction in vase life after storage was characterised by advanced signs of wilting on leaves and colour fading on petals. Increasing storage temperature from 1 to 10°C was strongly correlated with vase life decline for First Red' ($r^2 = 0.75$) and 'Akito' roses ($r^2 = 0.88$), respectively (Figure 4.2A, B, Appendix 4.1.1, Table A4.1.1.25. Duncan's multiple range tests indicated that vase lives of both cultivars stored at 10°C were significantly lower than that of control roses throughout the year (Appendix 4.1.1, Table A4.1.1.3 and A4.1.1.7).

The maximum potential quantum yield of PS II (dark adapted F_v/F_m) on day 0 of vase life was always greater for 'First Red' than for 'Akito' roses (Table 4.1). Growing season and storage temperature had significant effects on F_v/F_m (Appendix 4.1.1, Tables A4.1.1.2 and A4.1.1.6). Both cultivars when grown during winter had significantly lower F_v/F_m. The lower F_v/F_m during winter was due to reduction in F_v/F_m of stored flowers, while F_v/F_m of non-stored control roses remained close to the maximum value of 0.83 all year round (Figure 4.1C, D, Appendix 4.1.1, Tables A4.1.1.17 – A4.1.1.24). Thus, the reduction in F_v/F_m during winter is not due to direct effects of growing season but is due to an interactive effect of growing season and cold storage. F_v/F_m ratios declined with decreasing storage temperature (Table

4.1). Non-stored roses had the highest F_v/F_m. The decline in F_v/F_m of roses stored at 1°C was significantly (P<0.01) lower as compared to F_v/F_m value of control and roses stored at 5 and 10°C. However, according to Duncan's multiple range tests, this effect was only observed during winter experiments (Figure 4.1C, D, Appendix 4.1.1, Tables A1.5.1.4 and A4.1.1.8). The fall in F_v/F_m value for 'Akito' roses on day 0 was strongly correlated (r^2 = 0.97) with reduced storage temperature (Figure 4.3C, D, Appendix 4.1.1, Table A4.1.1.25). The slope of regression was less in 'First Red' roses (r^2 = 0.65).

Table 4.1: Effect of growing season and storage temperature on vase life (mean of flower and foliage lives) and F_v/F_m on d 0 of 'First Red' and 'Akito' roses. Flowers were grown from spring to winter and then were not stored (control) or stored wet at 1, 5 and 10°C for 10 days. Data are main-factor $\bar{x}$; n = 80. Data for independent treatment means each season are presented in Figure 4.1. ANOVA and Duncan's tests are presented in Appendix 4.1.1, Tables A4.1.1.1 – A4.1.1.8.

Main Factors	**'First Red'**		**'Akito'**	
	Vase life [a]	F_v/F_m	**Vase life**	F_v/F_m
1) Growing season [b]				
Spring	10.1 a	0.82 a	9.7 a	0.82 a
Summer	10.9 b	0.82 a	11.9 b	0.81 a
Autumn	7.4 c	0.82 a	9.0 a	0.80 a
Winter	6.0 d	0.75 b	7.4 c	0.68 b
2) Storage treatment				
Unstored	13.2 a	0.83 a	13.9 a	0.83 a
1°C	8.0 b	0.77 b	8.4 b	0.71 b
5°C	7.9 b	0.81 a	7.9 b	0.78 a
10°C	5.0 c	0.82 a	7.7 b	0.79 a

[a] Data are main factor means of vase life and F_v/F_m on d 0.

[b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

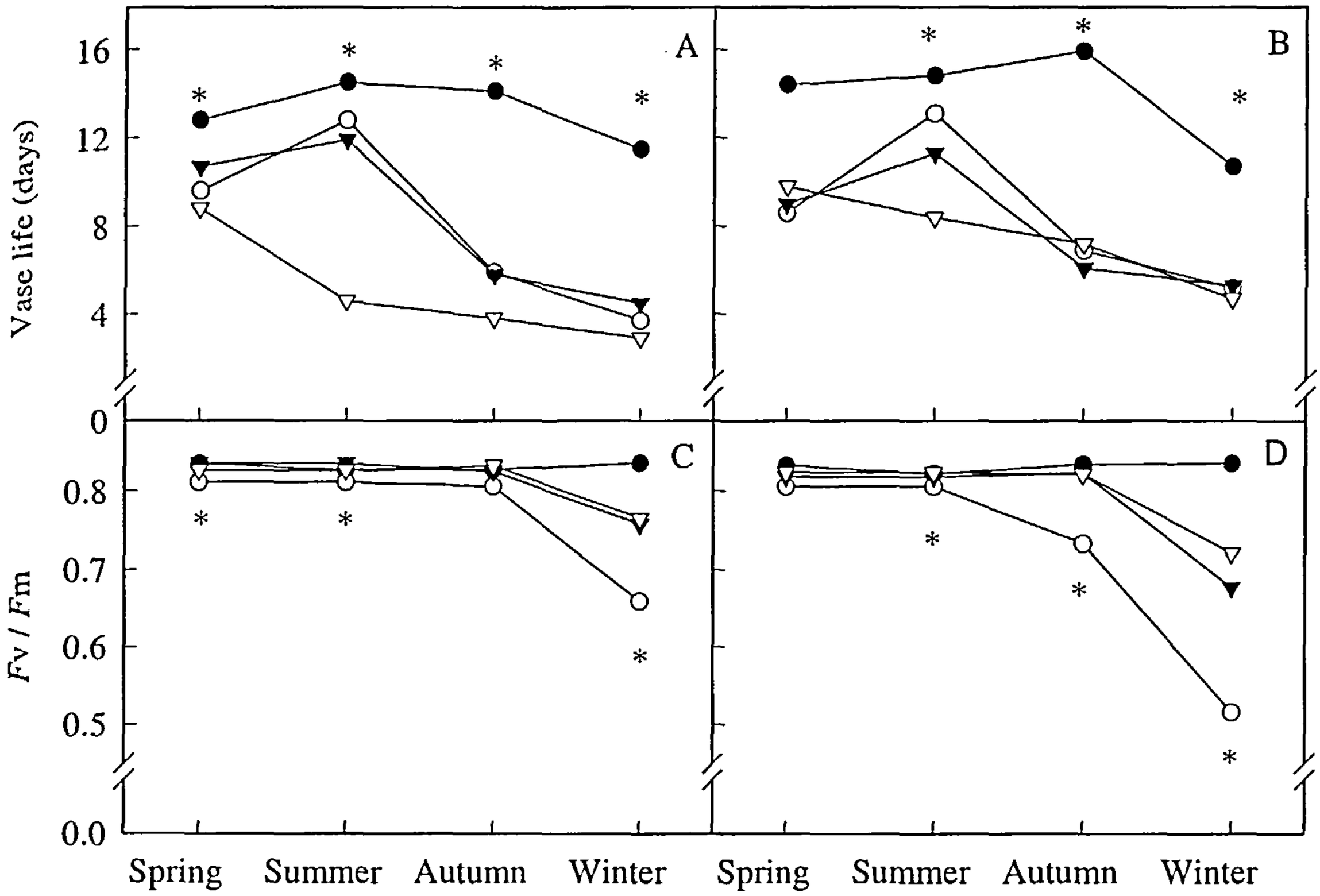


Figure 4.1: Changes in vase life (mean of flower and foliage lives) (A, B) and F_v/F_m on day 0 of vase life (C, D) for 'First Red' (A, C) and 'Akito' (B, D) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers during different growing seasons. Individual treatment data are $\bar{x}$; n = 20. Stars indicate significant difference between storage treatments for each season at P = 0.05 (Appendix 4.1.1, Tables A4.1.1.9 – A4.1.1.24).

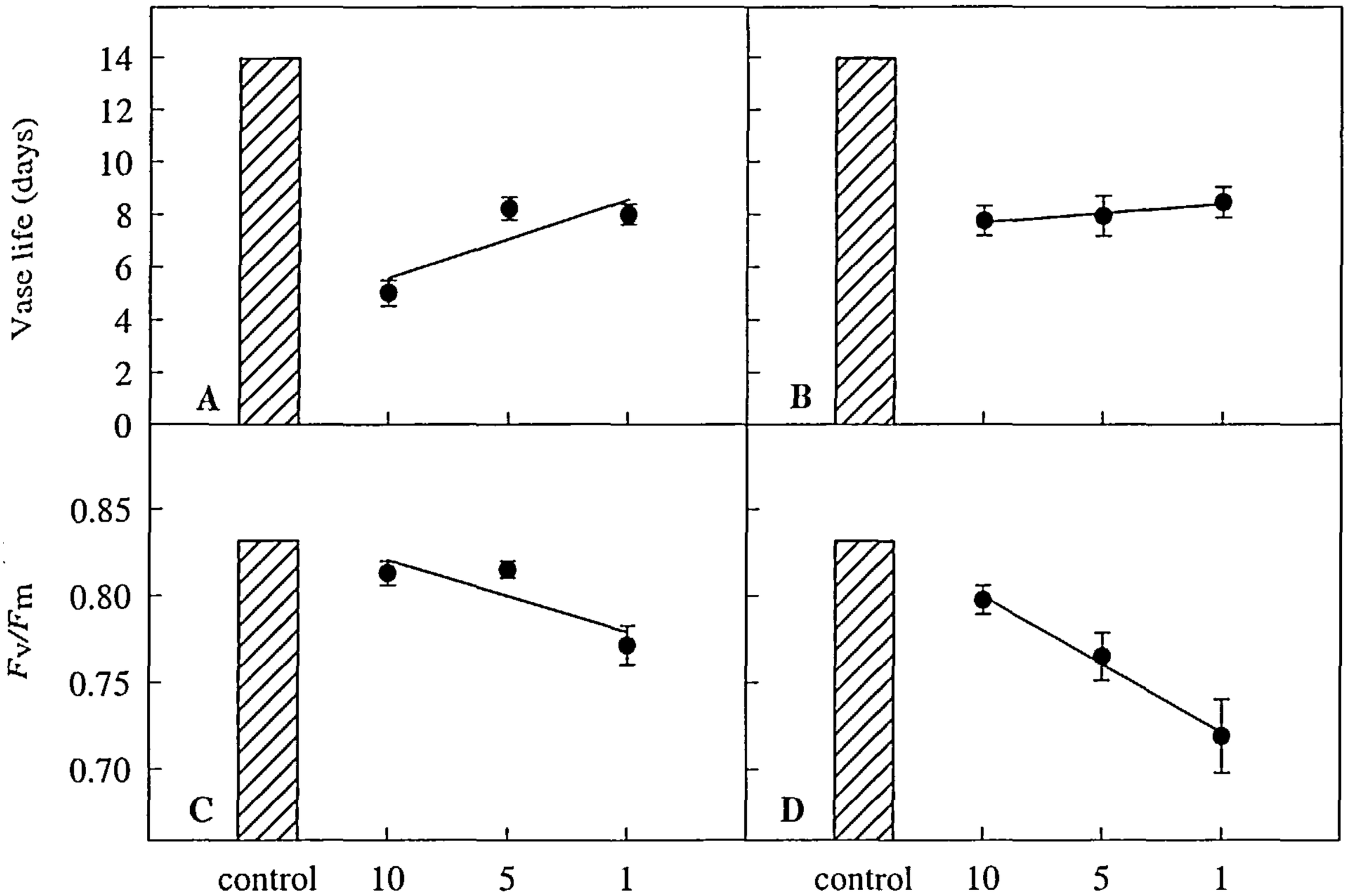


Figure 4.2: Simple linear regression analysis (y = a.x +/- b) between storage temperature (wet-stored flowers at 1, 5, and 10°C for 10 days) and vase life (A, B) or F_v/F_m values on day 0 for 'First Red' (A, C) and 'Akito' (B, D) roses. Histogram bars indicate vase lives and F_v/F_m values for non-stored control flowers. Vase life and F_v/F_m data are $\bar{x}$; n = 80 (20 replications per season for each storage treatment). Vertical bars on scatter plot show ± S.E. (n = 80) for each treatment. Regression parameters are presented in Appendix 4.1.1, Table A4.1.1.25. Regression lines were calculated from integer values but are presented as category values.

F_v/F_m values of the winter-grown flowers that were most sensitive to CI progressively reduced during vase life evaluation (Figure 4.3). The reduction in F_v/F_m was evident for both stored and non-stored control flowers. The capacity of 'First Red' and 'Akito' roses to maintain F_v/F_m decreased with reducing storage temperature from 10 to 1°C. This difference was more marked for 'Akito' versus

'First red' roses during vase life and consistently significant ($P \leq 0.05$) until d 8 (Appendix 4.1.1, Tables A4.1.1.26 – A4.1.1.35).

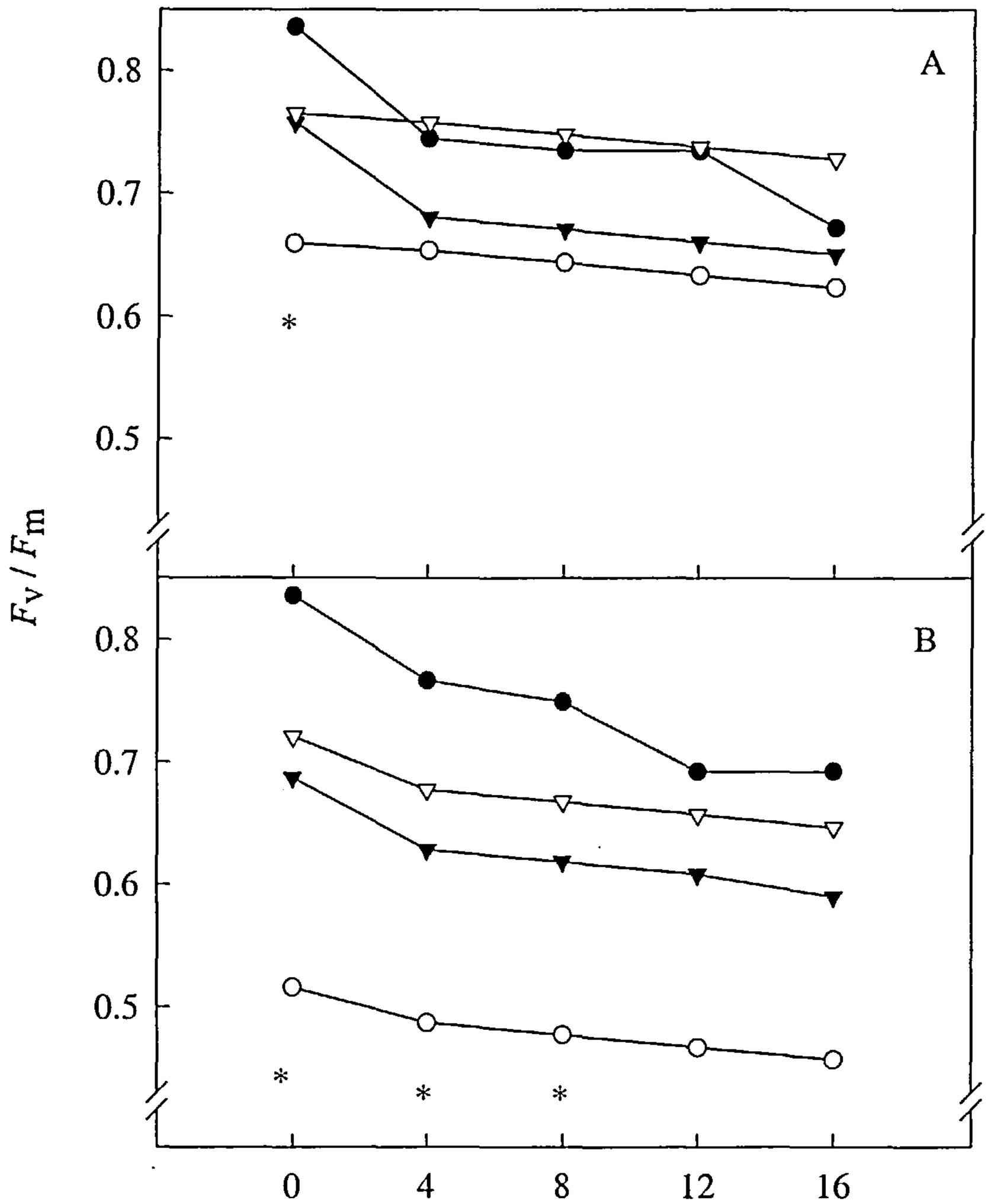


Figure 4.3: F_v/F_m changes during vase life for 'First Red' (A) and 'Akito' (B) roses grown during winter and following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers. Individual treatment data are $\bar{x}$; n = 20. Stars indicate significant difference between storage treatments for each day at P = 0.05 (Appendix 4.1.1, Table A4.1.1.26 – A4.1.1.35).

4.2.1.2 Flower condition

Flower stages

Flower opening averaged over storage treatments was greatest for roses grown during spring (Table 4.2, Appendix A4.1.2, Tables A4.1.2.1 – A4.1.2.8). In winter, 'First Red' and 'Akito' roses failed to open reaching a minimum flower stage of 2.27 on day 8 of vase life. Flower opening on day 8 was consistently greater for control than for stored flowers throughout the year (Figure 4.4A, B, Appendix 4.1.2, Tables A4.1.2.16 – A4.1.2.23). After storage at 1°C, 'First Red' and 'Akito' roses averaged flower stages of 2.02 and 2.70, respectively (Table 4.2). The reduced flower opening at 1°C was recorded from spring to summer for both cultivars.

Stored roses started their vase life 10 days (storage duration) after harvest while vase life for control flowers started at the day of harvest. Thus, both flower and foliage stages were more advanced for stored than for non-stored (freshly harvested) roses at the beginning of vase life (Figure 4.5, Appendix 4.1.3, Tables A4.1.3.1 – A4.1.3.20). After day 4, flower opening was consistently greater for control than for stored flowers (Figure 4.5A, B, Appendix 4.1.3, Tables A4.1.3.1 – A4.1.3.10). Flower opening was significantly reduced during vase life for roses stored at 1°C. The decreased flower opening at 1°C was more pronounced in 'First Red' roses, which did not pass stage 2 (early opening; Chapter 3, section 3.2.3.1). However, 'Akito' roses opened better even after storage at 1°C reaching stage 3 (mid opening). In the other three storage treatments (control, 5 and 10°C), 'Akito' flowers were at more advanced flower stages compared to 'First Red' throughout vase life.

Foliage stages

Growing season did not have significant effects ($P > 0.05$) on foliage stages measured on d 8 of vase life (Table 4.2, Appendix 4.1.2, Tables A4.1.2.9 – 4.1.2.15). However, storing roses at low temperature accelerated foliage wilting compared to control flowers. 'First Red' and 'Akito' roses stored at 1°C averaged minimum foliage stages of 2.02 and 2.70, respectively. Reduced foliage wilting of control flowers was recorded throughout the year for First Red' roses and from summer to

winter for 'Akito' roses (Figure 4.4C, D, Appendix 4.1.2, Tables A4.1.2.24 – A4.1.2.31).

Leaves of control flowers were upright with good turgor (Chapter 3, section 3.2.3.2) until d 8 of vase life, while leaves of stored flowers started to wilt directly after storage (Figure 4.5C, D, Appendix 4.1.3, Tables A4.1.3.11 – A4.1.3.20). The first signs of leaf softening for control flowers became visible after the day 8 when most leaves of stored flowers had already wilted. Leaves of stored roses had advanced signs of softening and wilting (stage 4) from day 4. Vase life ended for leaves of control flowers after day 12 (stage 4), when advanced signs of wilting and senescence were developed (personal observations).

Table 4.2: Effect of growing season and storage temperature on flower and foliage stages (scales 1-5) on d 8 of 'First Red' and 'Akito' roses. Flowers were grown from spring to winter and then were not stored (control) or stored wet at 1, 5 and 10°C for 10 days. Data are main-factor $\overline{x}$; n = 80. Data for independent treatment means each season are presented in Figure 4.4. ANOVA, non-parametric and Duncan's tests are presented in Appendix A4.1.2, Tables A4.1.2.1 – A4.1.2.15.

Main Factors	'First Red'		'Akito'	
	Flower stage [a]	Foliage stage	Flower stage	Foliage stage
1) Growing season [b]				
Spring	2.92 a	3.42 a	3.85 a	4.07 a
Summer	2.30 b	3.67 a	3.80 a	3.82 a
Autumn	2.55 b	3.50 a	3.25 b	3.87 a
Winter	2.27 b	3.52 a	2.27 c	3.77 a
2) Storage treatment				
Unstored	3.15 a	2.90 a	3.97 a	3.62 a
1°C	2.02 b	3.72 b	2.70 b	4.02 b
5°C	2.40 b	3.62 c	3.20 c	3.85 ab
10°C	2.47 b	3.87 c	3.30 c	4.05 b

[a] Data are main factor means of flower and foliage stages on d 8.

[b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

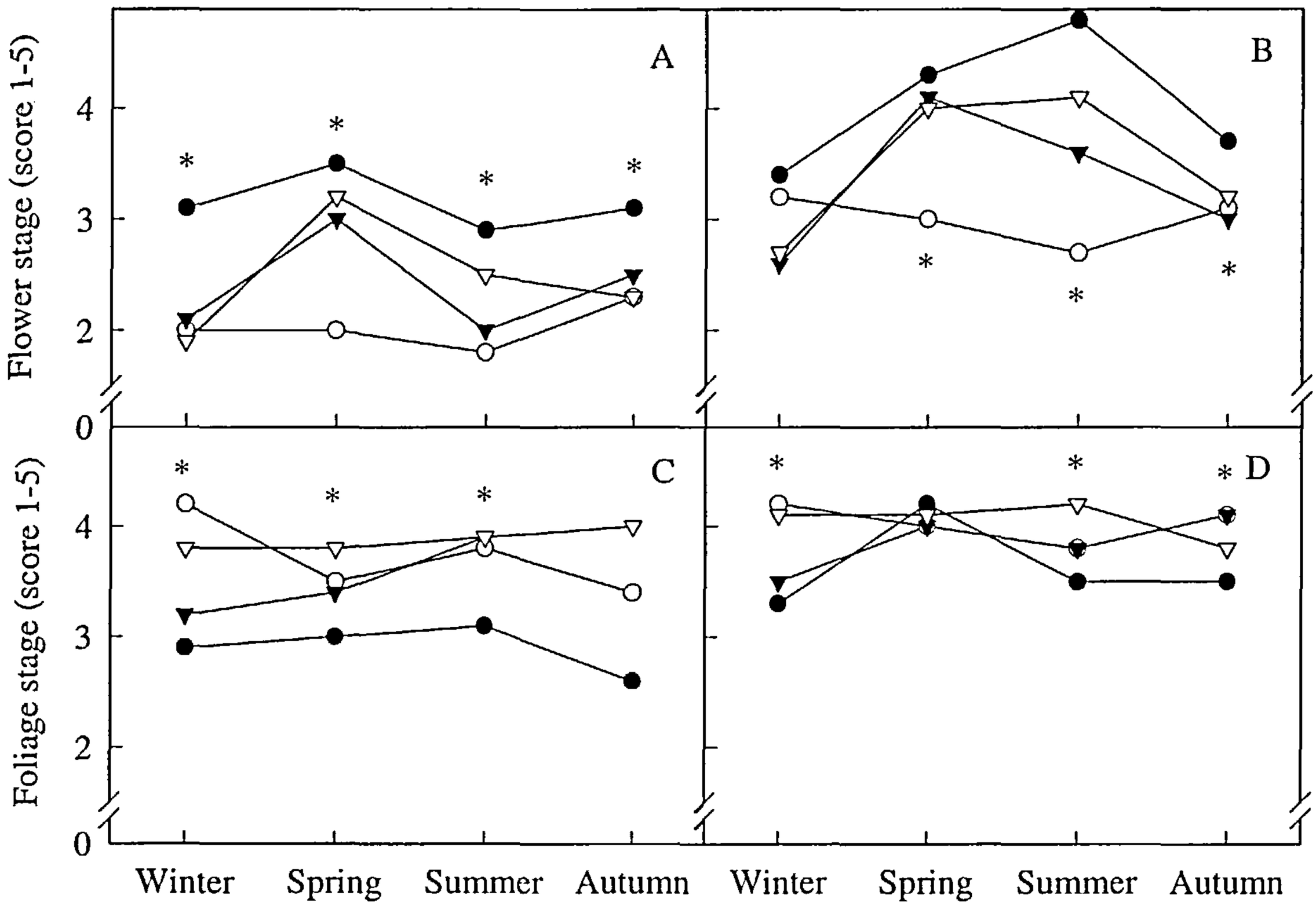


Figure 4.4: Changes in flower (A, B) and foliage (C, D) stages (scales 1-5) as measured on d 8 for 'First Red' (A, C) and 'Akito' (B, D) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers during different growing seasons. Individual treatment data are $\bar{x}$; $n = 20$. Stars indicate significant difference between storage treatments for each season at P = 0.05 (Appendix A4.1.2, Tables A4.1.2.16– A4.1.2.31).

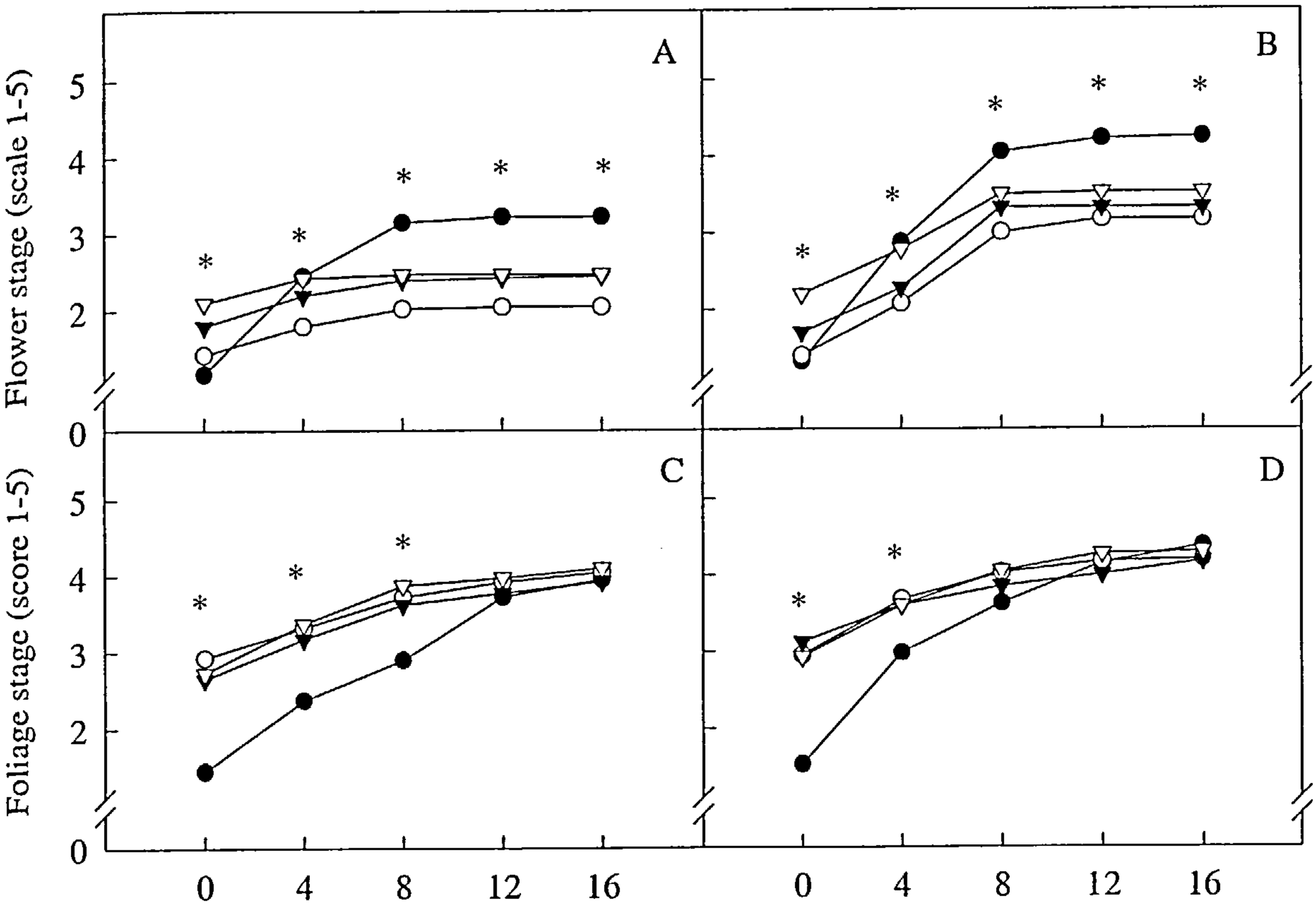


Figure 4.5: Changes in flower (A, B) and foliage (C, D) stages (scales 1-5) during vase life for 'First Red' (A, C) and 'Akito' (B, D) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers. Data are main factor means during the year; n = 80. Stars indicate significant difference between storage treatments for each day at P = 0.05 (Appendix 4.1.3, Tables A4.1.3.1 – A4.1.3.20).

Petal colour

Decline in *b** coefficient indicates the developed blueing in red cultivars (Papadimitriou, 1995), such as 'First Red' petals. *b** value of 'First Red' petals was significantly ($P < 0.05$) lower when roses were grown from autumn to winter, indicating advanced signs of petal blueing (Table 4.3, Appendix 4.1.4, Tables A4.1.4.1 and A4.1.4.2). Control roses averaged a *b** value of 11.09 while those stored at 1, 5 and 10°C averaged *b** values of 7.55, 6.54 and 5.37, respectively. The relatively increased *b** value of control roses from spring to summer (Figure 4.6, Appendix 4.1.4, Table A4.1.4.3 – A4.1.4.6) indicates less petal blueing. In summer, the lower *b** value of 'First Red' roses stored at 10°C was significant different compared to control and stored roses at 1 and 5°C (Appendix 4.1.4, Table A4.1.4.5).

Table 4.3: Effect of growing season and storage temperature on *b** value of 'First Red' petals on d 8. Flowers were grown from spring to winter and then were not stored (control) or stored wet at 1, 5 and 10°C for 10 days. Data are main-factor $\bar{x}$; n = 80. Data for independent treatment means are presented in Figure 4.6. ANOVA and Duncan's tests are presented in Appendix 4.1.4, Tables A4.1.4.1 and A4.1.4.2.

Main Factors	**b* value** [b]
1) Growing seasons [a]	
Spring	11.34 a
Summer	11.49 a
Autumn	3.26 b
Winter	4.46 b
2) Storage treatments	
Unstored	11.09 a
1°C	7.55 b
5°C	6.54 bc
10°C	5.37 c

[a] Data are main factor means of *b** value on petals on d 8.

[b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

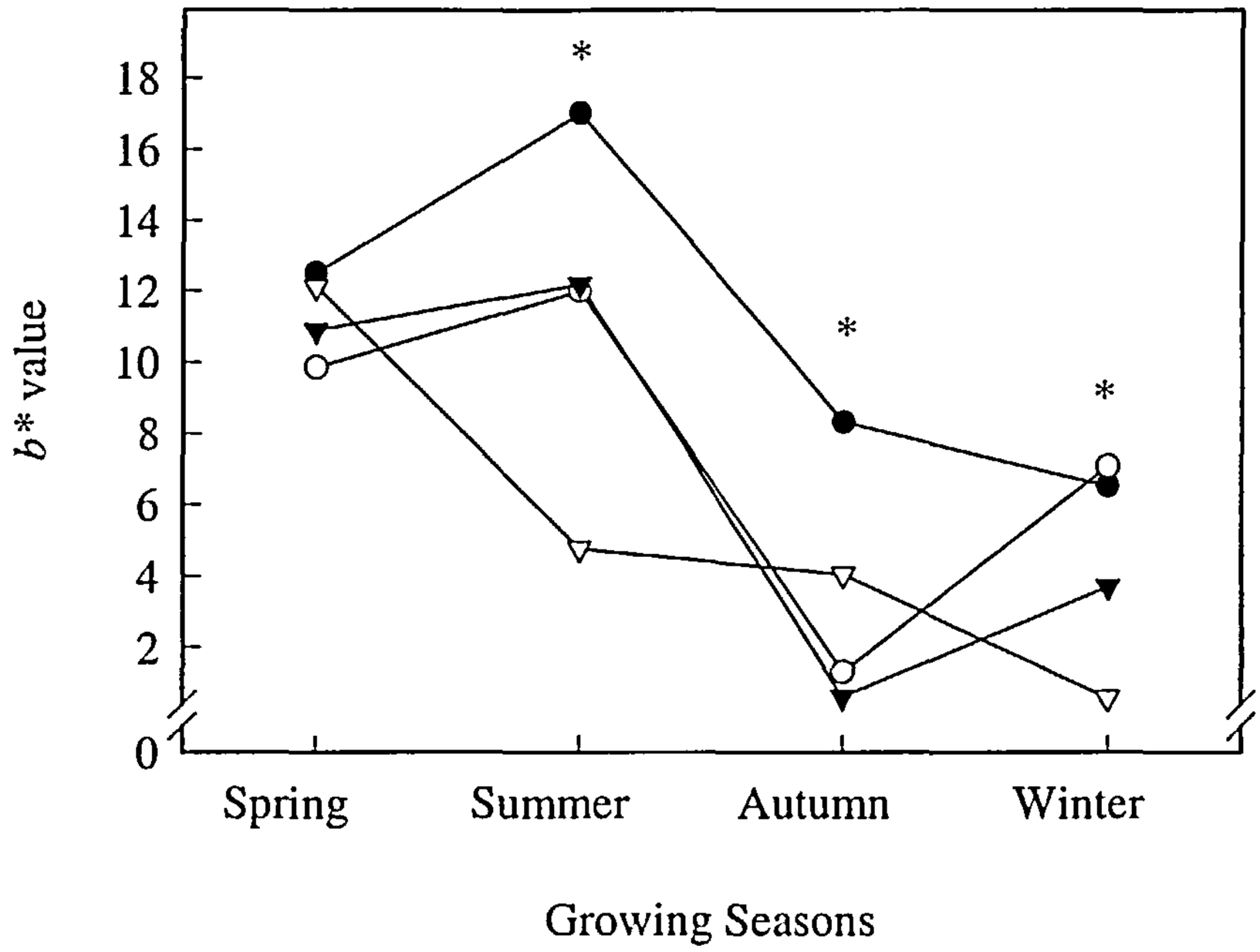


Figure 4.6: Changes in *b** value on d 8 for 'First Red' roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers during different growing seasons. Individual treatment data are $\bar{x}$; n = 20. Stars indicate significant difference between storage treatments for each season at P = 0.05 (Appendix 4.1.4, Table A4.1.4.3 – A4.1.4.6).

*b** coefficient of flower petals progressively decreased for 'First Red' roses during vase life (Figure 4.7, Appendix 4.1.4, Table A4.1.4.7 – A4.1.4.11). On d 0 of vase life, blueing was significantly (P < 0.05) greater (e.g. lower *b** value) for stored roses comparing to controls. Although *b** coefficient fell sharply for controls after d 0, it was maintained higher than that of stored roses. Storage of 'First Red' flowers at 10°C caused more blueing on petals after day 4 of vase life, followed by storage at 5 and 1°C.

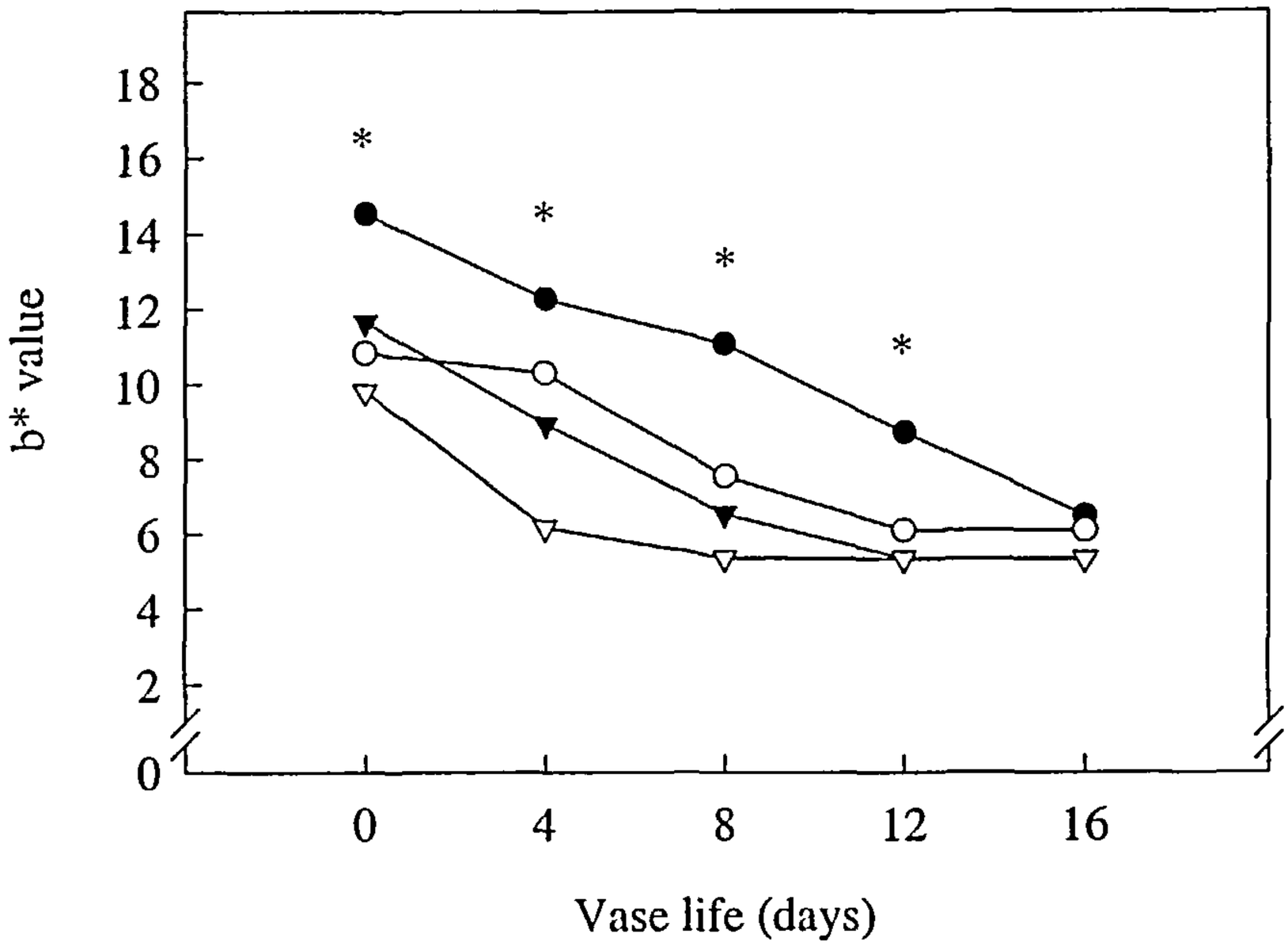


Figure 4.7: Changes in *b** value during vase life for 'First Red' petals following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers. Data are main factor means during the year; n = 80. Stars indicate significant difference between storage treatments for each day at P = 0.05 (Appendix 4.1.4, Table A4.1.4.7 – A4.1.4.11).

Bent neck

Under the conditions of current experiments, 'First Red' roses did not show bent neck symptoms (Table 4.4, Appendix 4.1.5, Table A4.1.5.1). On the other hand, 'Akito' roses were most sensitive to bent neck during the year. However, this increased sensitivity of 'Akito' roses to bent neck did not result in vase life losses, as the symptom was recorded on the last days of vase life (personal observation). 'Akito' flowers grown in spring had significantly ($P < 0.05$) highest sensitivity to bent neck followed by those grown in winter, autumn and summer (Appendix 4.1.5, Table A4.1.5.2). Bent neck incidence of 'Akito' roses was about equal for control and stored roses at 1 and 5°C (Table 4.13, Appendix 4.5.5, Tables A4.5.5.2) while flowers stored at 10°C had less bent necks.

Table 4.4: Bent neck incidence (%) of 'First Red' and 'Akito' roses grown from spring to winter and then stored wet at 1, 5 and 10°C or were not stored (control). Individual treatment data are $\bar{x}$; $n = 20$. ANOVA tables are presented in Appendix 4.1.5, Tables A4.1.5.1 and 4.1.5.2.

Storage treatment	Growing seasons	Bent neck (%) [a]	
		'First Red'	'Akito'
Control	Spring	0.0a	40.0a, b
1°C		0.0a	40.0a, b
5°C		0.0a	80.0a
10°C		0.0a	0.0b
Column means		0.0	40.0
Control	Summer	0.0a	0.0a
1°C		0.0a	0.0a
5°C		0.0a	40.0b
10°C		0.0a	0.0a
Column means		0.0	10.0
Control	Autumn	0.0a	40.0a
1°C		0.0a	0.0a
5°C		0.0a	20.0a
10°C		20.0a	0.0a
Column means		5.0	15.0
Control	Winter	0.0a	20.0a
1°C		0.0a	40.0a
5°C		0.0a	0.0a
10°C		0.0a	20.0a
Column means		0.0	20.0

[a] Within growing seasons, numbers followed by the same letter are not significantly different at P = 0.05.

4.2.1.3 Solution usage and fresh weight

Neither growing seasons nor storage treatments affected significantly vase solution usage by 'First Red' roses at the middle of vase life (day 7) (Table 4.5, Figure 4.8A, Appendix 4.1.6, Table A4.1.6). In 'First Red' roses, solution usage on day 7 was lowest by flowers stored at 10°C in summer and highest by control flowers in winter (Figure 4.8A, Appendix 4.1.6, Tables A4.1.6.8 – A4.1.6.11). In 'Akito' roses, flowers grown in autumn and spring had highest solution usage followed by those grown in summer and winter. Moreover, from autumn to spring solution usage by control was comparatively higher than by stored flowers (Figure 4.8B, Appendix 4.1.6, Tables A4.1.6.12 – A4.1.6.15). In winter experiments, increased solution usage by control was significant compared to stored roses (Figure 4.8B, Appendix 4.1.6, Table A4.1.6.3). In spring experiments, however, increased solution usage by control was significant only compared to roses stored at 10°C. Solution usage by 'Akito' roses was generally greater than that by 'First Red' at all storage treatments throughout the year (Figure 4.8A, B). Both cultivars had lowest solution usage after storage at 10°C but this difference was not always significant.

Fresh weight on day 8 averaged over storage treatments was best maintained for roses grown during summer (Table 4.5, Appendix 4.1.6, Tables A4.1.6.4 – A4.1.6.7). Highest losses of fresh weight were recorded in winter, while fresh weight of roses grown in spring and autumn was intermediate. Fresh weight maintenance was consistently greater for control than for stored roses throughout the year (Figure 4.8C, D). Greatest losses of fresh weight on day 8 were recorded for roses stored at 10°C. This loss of fresh weight at 10°C was observed throughout the year and from winter to summer in 'Akito' and 'First Red' roses, respectively (Appendix 4.1.6, Tables A4.1.6.16 – A4.1.6.23). 'Akito' had generally greater capacity in maintaining fresh mass than 'First Red' roses at all storage treatments during the year.

Relative fresh weights generally increased during the first days of vase life for non-stored control roses (Figure 4.9A, B, Appendix 4.1.7, Tables A4.1.7.1 – A4.1.7.16). The great maintenance of fresh weights by controls was consistently significant ($P < 0.05$) comparing to stored roses for both cultivars throughout vase life. Both 'First Red' and 'Akito' roses had significantly ($P < 0.05$) less capacity in maintaining fresh weight throughout vase life when they were stored at 10°C. Fresh weight of 'Akito' roses was generally better maintained than that of 'First Red'. In

'Akito' roses, fresh weights of controls were largely maintained > 100% of initial weight until day 8. In 'First Red' roses, fresh weight of controls was increased until day 6 and then decreased sharply until the end of vase life. Fresh weights of stored flowers decreased from day 0 except from 'Akito' roses stored at 1°C, which tended to maintain their fresh mass until day 4.

Table 4.5: Effect of growing season and storage temperature on solution usage (ml g^{-1} initial f.w.) on d 7 and fresh weight (% of initial f.w.) on d 8 of 'First Red' and 'Akito' roses. Flowers were grown from spring to winter and then were not stored (control) or stored wet at 1, 5 and 10°C for 10 days. Data are main-factor $\bar{x}$; n = 80. Data for independent treatment means are presented in Figure 4.8. ANOVA and Duncan's tests are presented in Appendix 4.1.6, Tables A4.1.6.1 – A4.1.6.7.

Main Factors	**'First Red'**		**'Akito'**	
	Solution usage [a]	**Fresh weight**	**Solution usage**	**Fresh weight**
1) Growing season [b]				
Spring	0.19 a	81.4 ab	0.45 a	84.3 a
Summer	0.13 a	85.4 b	0.33 b	88.9 a
Autumn	0.32 a	79.8 a	0.65 c	91.3 a
Winter	0.13 a	53.1 c	0.23 d	72.8 b
2) Storage treatment				
Unstored	0.27 a	86.5 a	0.60 a	100.2 a
1°C	0.16 a	73.5 b	0.41 c	82.4 b
5°C	0.26 a	73.5 b	0.36 bc	84.2 b
10°C	0.08 a	66.1 c	0.29 b	70.51 c

[a] Data are main-factor means of solution usage on d 7 and fresh weight on d 8.

[b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

Vase solution usage by flowers progressively decreased during vase life, indicating lower rates of water absorption by rose stems at the end of vase life (Figure 4.9C, D, Appendix 4.1.7, Tables A4.1.7.17 – A4.1.7.32). Solution usage by 'Akito' roses was consistently significant than that by 'First Red' throughout vase life. In

'First Red' roses, solution usage by controls was slightly greater compared to stored flowers (Figure 4.9C, Appendix 4.1.7, Tables A4.1.7.17 – A4.1.7.24). However, in 'Akito' roses solution usage by controls was significantly ($P < 0.05$) greater than that by stored flowers throughout vase life (Figure 4.9D, Appendix 4.1.7, Tables A4.1.7.25 – A4.1.7.32). Storing both cultivars at 10°C resulted in lowest solution usage during vase life.

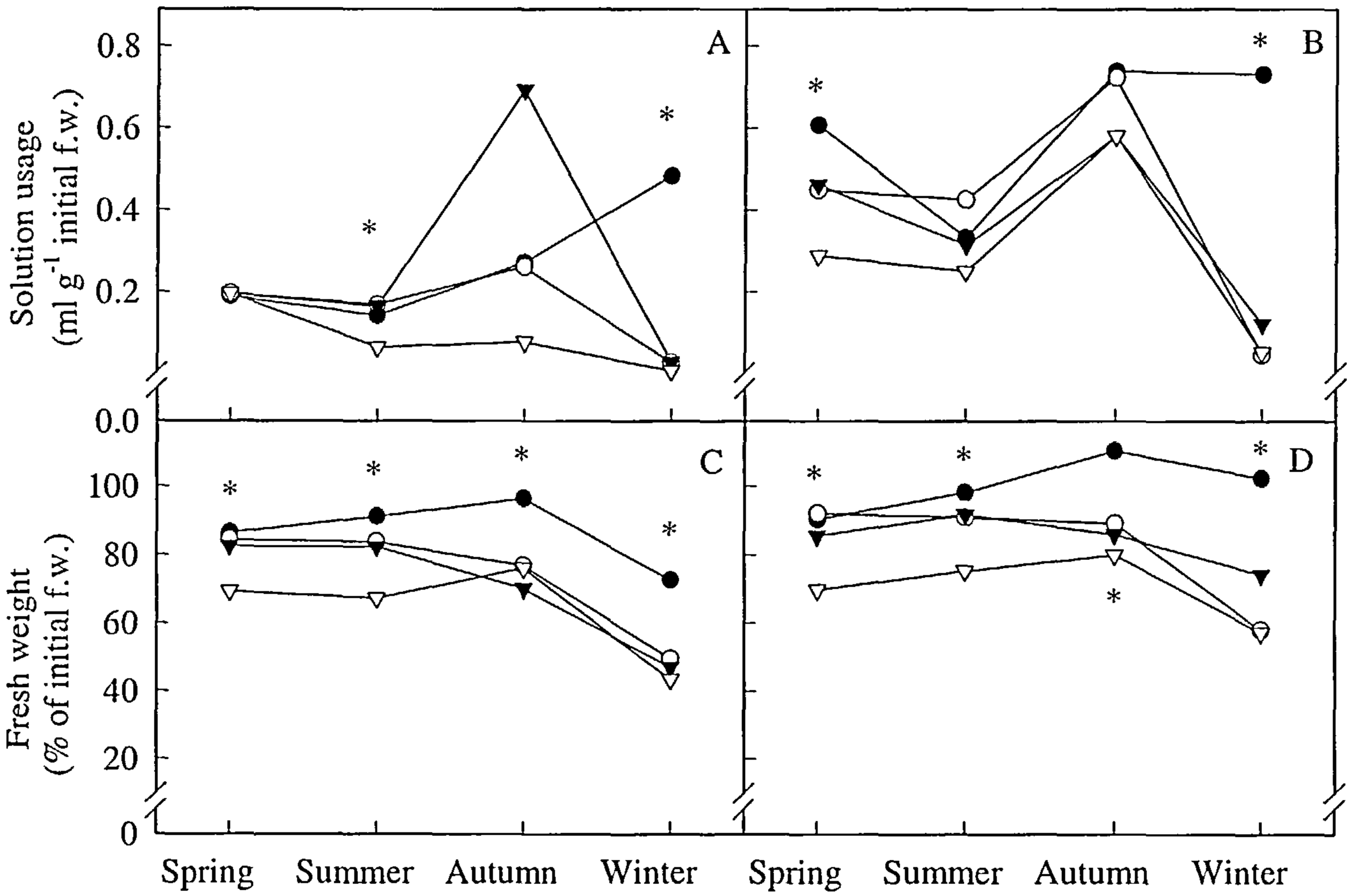


Figure 4.8: Changes in solution usage (A, B) on d 7 and relative fresh weight (C, D) on d 8 for 'First Red' (A, C) and 'Akito' (B, D) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers during different growing seasons. Individual treatment data are $\bar{x}$; $n = 20$. Stars indicate significant difference between storage treatments for each season at $P = 0.05$ (Appendix A4.1.6, Tables A4.1.6.8 – A4.1.6.23).

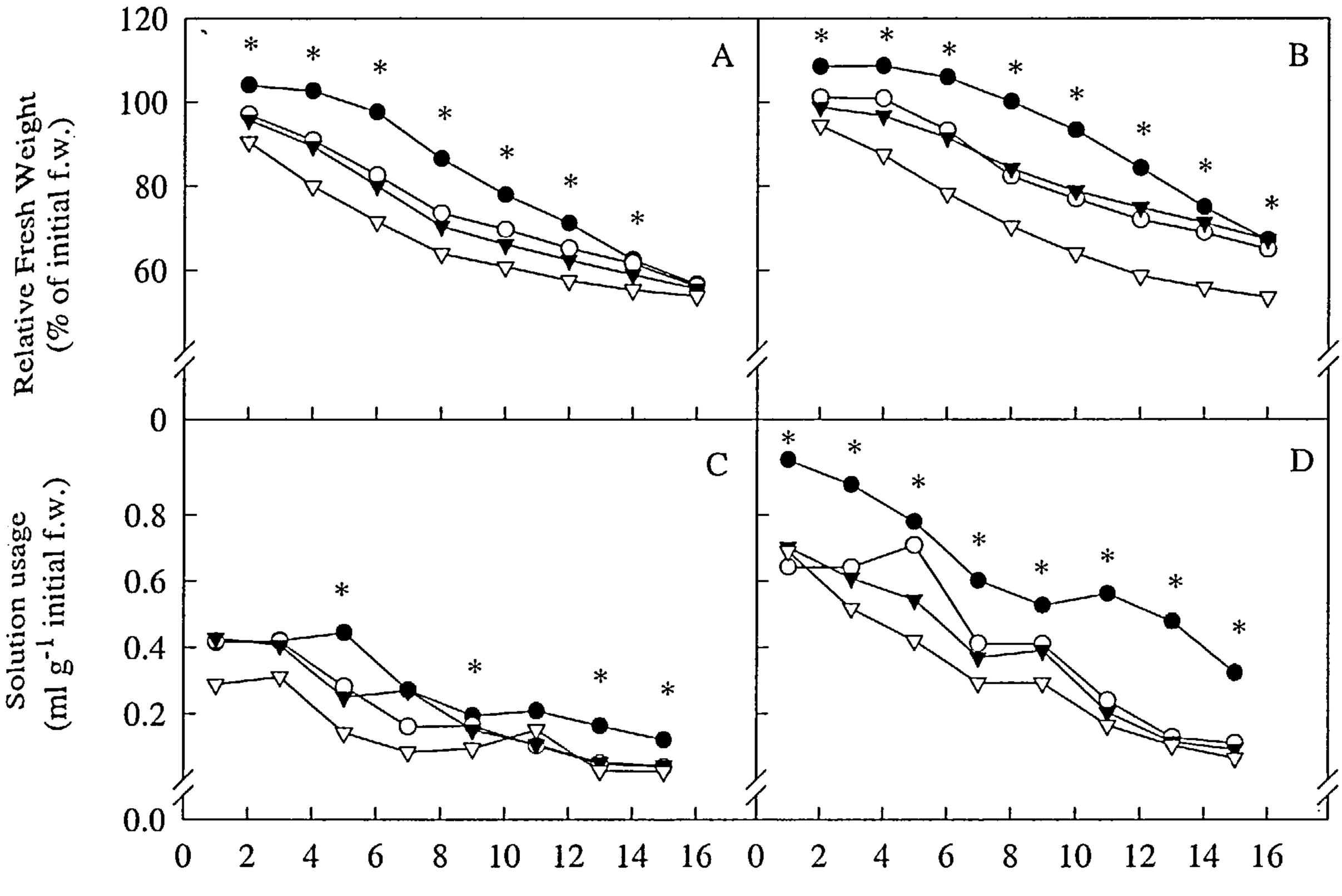


Figure 4.9: Changes in fresh weight (A, B) and solution usage (C, D) during vase life by 'First Red' (A, C) and 'Akito' (B, D) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers. Data are main factor means during the year; n = 80. Stars indicate significant difference between storage treatments for each day at P = 0.05 (Appendix 4.1.7).

4.2.1.4 Vase solution parameters

Final pH

Growing season and storage treatments significantly affected final pH of 'First Red' roses (Table 4.6, Appendix 4.1.8, Tables A4.1.8.1 and A4.1.8.2). In 'Akito' roses, final pH was affected only by storage treatment (Appendix 4.1.8, Tables A4.1.8.3 and A4.1.8.4). Both ANOVA and Duncan's multiple range tests showed that solution pH of 'Akito' roses at the end of vase life was consistently ($P < 0.05$) greater for control than for stored roses throughout the year (Figure 4.10B, Appendix 4.1.8, Tables 4.1.8.4 and A4.1.8.9 – A4.1.8.12). In 'First Red' roses, this difference was significant only in spring and autumn experiments (Figure 4.10A, Appendix 4.1.8, Tables 4.1.8.2 and A4.1.8.5 – A4.1.8.8).

Table 4.6: Effect of growing season and storage temperature on final pH of 'First Red' and 'Akito' roses. Flowers were grown from spring to winter and then were stored wet at 1, 5 and 10°C or not stored (control). Data are main-factor $\bar{x}$; n = 80. Data for independent treatment means are presented in Figure 4.10. ANOVA and Duncan's tests are presented in Appendix 4.1.8, Tables A4.1.8.1 and A4.1.8.4.

Main Factors	**Final pH** [a]	
	'First Red'	**'Akito'**
1) Growing season [b]		
Spring	3.61 b	3.61 a
Summer	3.70 ab	3.70 b
Autumn	3.83 ac	3.67 ab
Winter	3.91 c	3.65 ab
2) Storage treatment		
Unstored	4.10 a	3.95 a
1°C	3.67 b	3.58 bc
5°C	3.60 b	3.50 b
10°C	3.68 b	3.60 c

[a] Data are main factor means of final pH. [b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

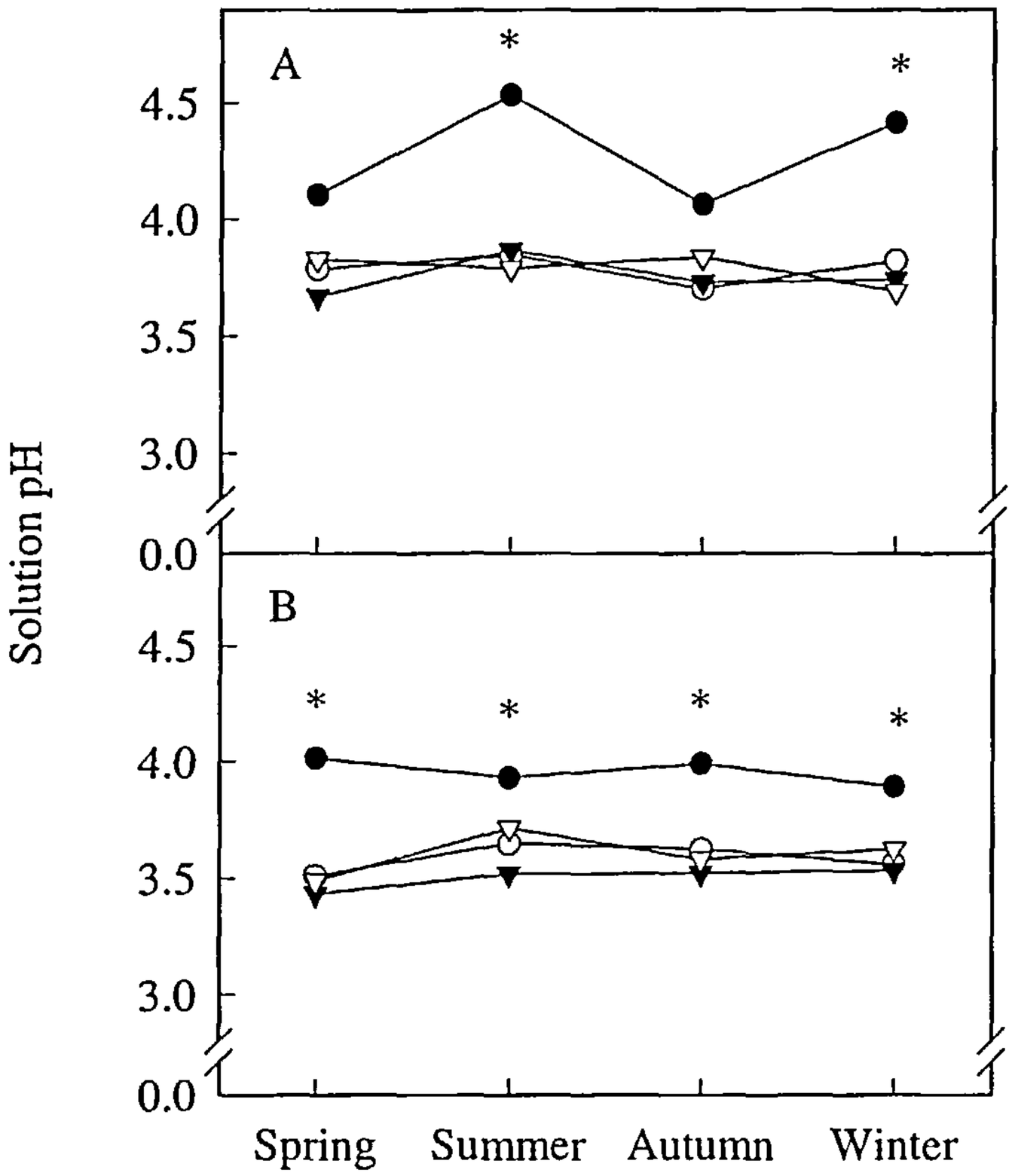


Figure 4.10: Changes in final pH of vase solution for 'First Red' (A) and 'Akito' (B) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers during different growing seasons. Individual treatment data are $\bar{x}$; $n = 20$. Stars indicate significant difference between storage treatments for each season at P = 0.05 (Appendix 4.1.8, Tables A4.1.8.5 – A4.1.8.12).

Absorbance and solution turbidity

Turbidity scores and vase solution absorbance at the end of experiments showed that microbial growth was not a problem for roses grown from autumn to winter (Table 4.7, Appendix 4.1.9, Tables A4.1.9.1 – A4.1.9.7). The relative absence of absorbance was recorded for both control and stored roses (Figure 4.11A, B, Appendix 4.1.9, Tables A4.1.9.4 and A4.1.9.8 – A4.1.9.15). Vase solutions for roses

grown from spring to summer were most turbid, indicating more microbial growth in the vase. In Akito' roses, solution turbidity was consistently greater for control than for stored flowers throughout the year. This reproducible effect of storage, which was significant ($P < 0.05$) in summer, may be due to suppression of microbial growth at low temperature (Figure 4.11C, Appendix 4.1.9, Tables A4.1.9.20 – A4.1.9.23). In 'First Red' roses, however, increased solution turbidity of control flowers was only recorded from summer to autumn (Figure 4.11D, Appendix 4.1.9, Tables A4.1.9.16 – A4.1.9.19).

Table 4.7: Effect of growing season and storage temperature on absorbance (OD's at 400, 500 and 600 nm) and solution turbidity (scale 1-3) of 'First Red' and 'Akito' roses at the end of vase life. Flowers were grown from spring to winter and then were stored wet at 1, 5 and 10°C or not stored (control). Data are main-factor $\bar{x}$; n = 80. Data for independent treatment means are presented in Figure 4.11. ANOVA and Duncan's tests are presented in Appendix 4.1.9, Tables 4.1.9.1 – 4.1.9.7.

Main Factors	'First Red'		'Akito'	
	Absorbance [a]	Turbidity	Absorbance	Turbidity
1) Growing season [b]				
Spring	0.034 a	1.40 ab	0.026 ab	1.50 a
Summer	0.040 a	1.67 a	0.028 b	1.35 ab
Autumn	0.006 b	1.35 b	0.002 c	1.15 b
Winter	0.018 c	1.17 b	0.020 a	1.22 ab
2) Storage treatment				
Unstored	0.024 a	1.65 a	0.021 a	1.67 a
1°C	0.026 a	1.27 bc	0.018 a	1.12 b
5°C	0.023 a	1.17 c	0.019 a	1.17 b
10°C	0.026 a	1.50 ab	0.018 a	1.25 b

[a] Data are main factor means of absorbance and turbidity score.

[b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

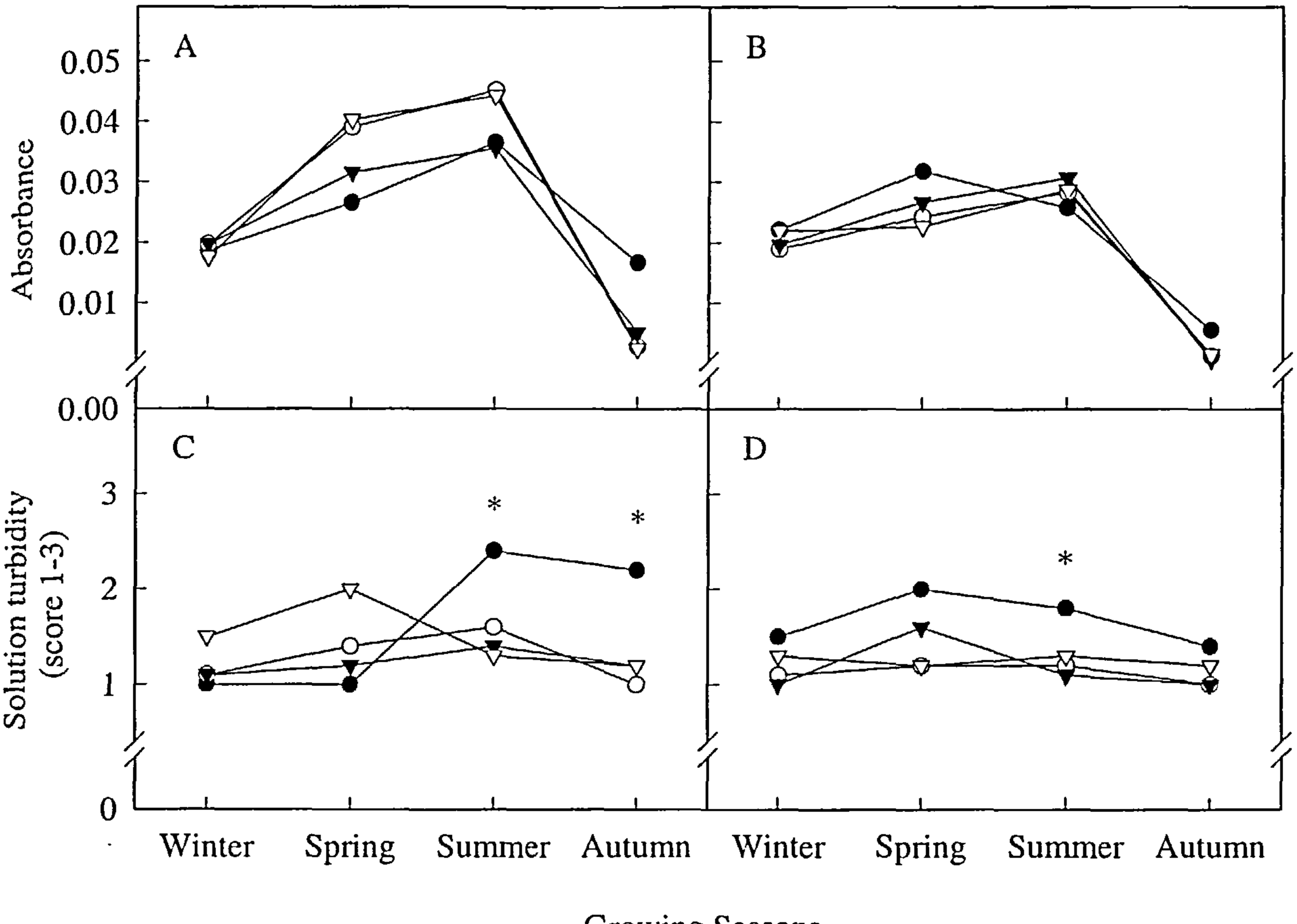


Figure 4.11: Changes in absorbance and solution turbidity (score 1-3) of vase solution for 'First Red' (A, C) and 'Akito' (B, D) roses following wet storage at 1 (○), 5 (▼), and 10°C (▽) for 10 days and for the non-stored control (●) flowers during different growing seasons. Individual treatment data are $\bar{x}$; $n = 20$. Stars indicate significant difference between storage treatments for each season at $P = 0.05$ (Appendix 4.1.9, Tables A4.1.9.8 – A4.1.9.23).

4.2.1.5 Relationship between vase life duration and vase life parameters

Vase life duration was significantly ($P < 0.01$) correlated with F_v/F_m on d 0 and flower stage on d 8 (Table 4.8). These weak but significant correlations indicate that increases in F_v/F_m at the beginning and flower stage in the middle of vase life are associated with vase life extension. Greatest and significant ($P < 0.01$) correlations were also found for 'First Red' ($r^2 = 0.73$) and 'Akito' roses ($r^2 = 0.62$) between vase lives and fresh weights on d 8. This positive correlation shows that fresh weight at the middle of vase life is a good indication of vase life duration. Moreover, in 'Akito' roses, solution usage by flowers was significantly correlated with vase life. Finally, a significant ($P < 0.01$) but negative correlation between vase life duration and foliage stages was found for 'First Red' ($r^2 = -0.48$) and 'Akito' ($r^2 = -0.35$) flowers. This negative correlation indicates that advanced foliage stages are strongly associated with vase life decline.

Table 4.8: Effect of vase life duration on vase life parameters (e.g. F_v/F_m on d 0, flower and foliage stages on d 8, fresh weight on d 8 and solution usage on d 7) of 'First Red' and 'Akito' roses grown from autumn 2002 to summer 2003 and then put directly in the vases (control) or stored at 1, 5 and 10°C for 10 days.

Vase life variables	**Number of replications**	**Vase life duration** [a]	
		'First Red'	**'Akito'**
F_v/F_m (d 0)	320	0.43 **	0.44 **
Flower stage (d 8)	320	0.38 **	0.50 **
Foliage stage (d 8)	320	-0.48 **	-0.35 **
Fresh weight (d 8)	320	0.73 **	0.69 **
Solution usage (d 7)	320	0.20 n.s.	0.47 **

[a] Data are results from Pearson's correlation at P = 0.05.

** Significance at P = 0.01, n.s. not significant at P = 0.05.

4.2.1.6 Water loss and dry weight of detached leaves

Growing season, temperature and relative humidity (RH) significantly affected water loss of leaves detached from 'First Red' and 'Akito' roses at the time of harvest (Table 4.9, Appendix 4.2.1, Table A4.2.1.1). As a main factor mean for all growing seasons, temperatures and relative humidity levels, leaves of 'Akito' averaged a water loss of 30% and those of 'First Red' flowers averaged a water loss of 22%. Increased water loss was recorded in autumn for both cultivars compared to winter, spring and summer (Figure 4.12). Detached leaves of roses grown in autumn had the greatest water loss of 47.4% followed by those grown in summer with losses of 21% and those grown in winter and spring with 18.5 and 17.2%, respectively. This significant increase of water loss during autumn was recorded at all temperature and RH levels for both cultivars (Figure 4.12). Increasing storage temperature from 1 to 10°C resulted in water loss increase by detached leaves of 'Akito' roses (Figure 4.12B, D, F, Appendix 4.2.1, Tables A4.2.1.14 – A4.2:1.25). In 'First Red' roses, however, leaves at 10°C had greater water losses than that at 1 and 5°C in winter and spring experiments. This difference was marked at all RH levels (Figure 4.12A, C, E, Appendix 4.2.1, Tables A4.2.1.2 – A4.2.1.13). Increasing RH from 75 to 95% resulted in water loss decline, indicating less evaporation at increased RH levels. This clear effect of RH was recorded for both cultivars throughout the year.

Detached leaves of roses grown in spring averaged greatest dry weights of 0.49 g followed by those grown in summer, winter and autumn with dry weights of 0.46, 0.44 and 0.41 g, respectively (Table 4.9, Appendix 4.2.2, Table A4.2.2.1). This significant increase ($P < 0.05$) in dry weights of leaves grown in spring was recorded for both cultivars at all temperature and RH levels (Figure 4.13, Appendix 4.2.2, Tables A4.2.2.2 – A4.2.2.25).

Table 4.9: Effect of season, storage temperature, RH and cultivar on water loss and dry weight of detached leaves from rose stems. Data are means of water loss measured 12, 24 and 48 h after detachment. Data for independent treatment means are presented in Figures 4.12 – 4.13 and ANOVA in Appendix 4.2, Tables A4.2.1.1 and A4.2.2.1.

Factors	Number of samples [b]	Water loss (%) [a]	Dry weight (g)
1) Growing season [c]			
Autumn	180	47.4 a	0.41 a
Winter	180	18.5 b	0.44 b
Spring	180	17.2 b	0.49 c
Summer	180	21.0 c	0.46 b
2) Temperature (°C)			
1	240	22.3 a	0.45 a
5	240	26.5 b	0.44 a
10	240	29.3 c	0.45 a
3) Relative humidity(%)			
75	240	31.0 a	0.46 a
85	240	26.3 b	0.46 a
95	240	20.7 c	0.43 b
4) Cultivar			
'First Red'	360	22.0 a	0.56 a
'Akito'	360	30.0 b	0.33 b

[a] Data are means of water loss measured 12, 24 and 48 h after detachment

[b] Data are main factor means of water loss and dry weight.

[c] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

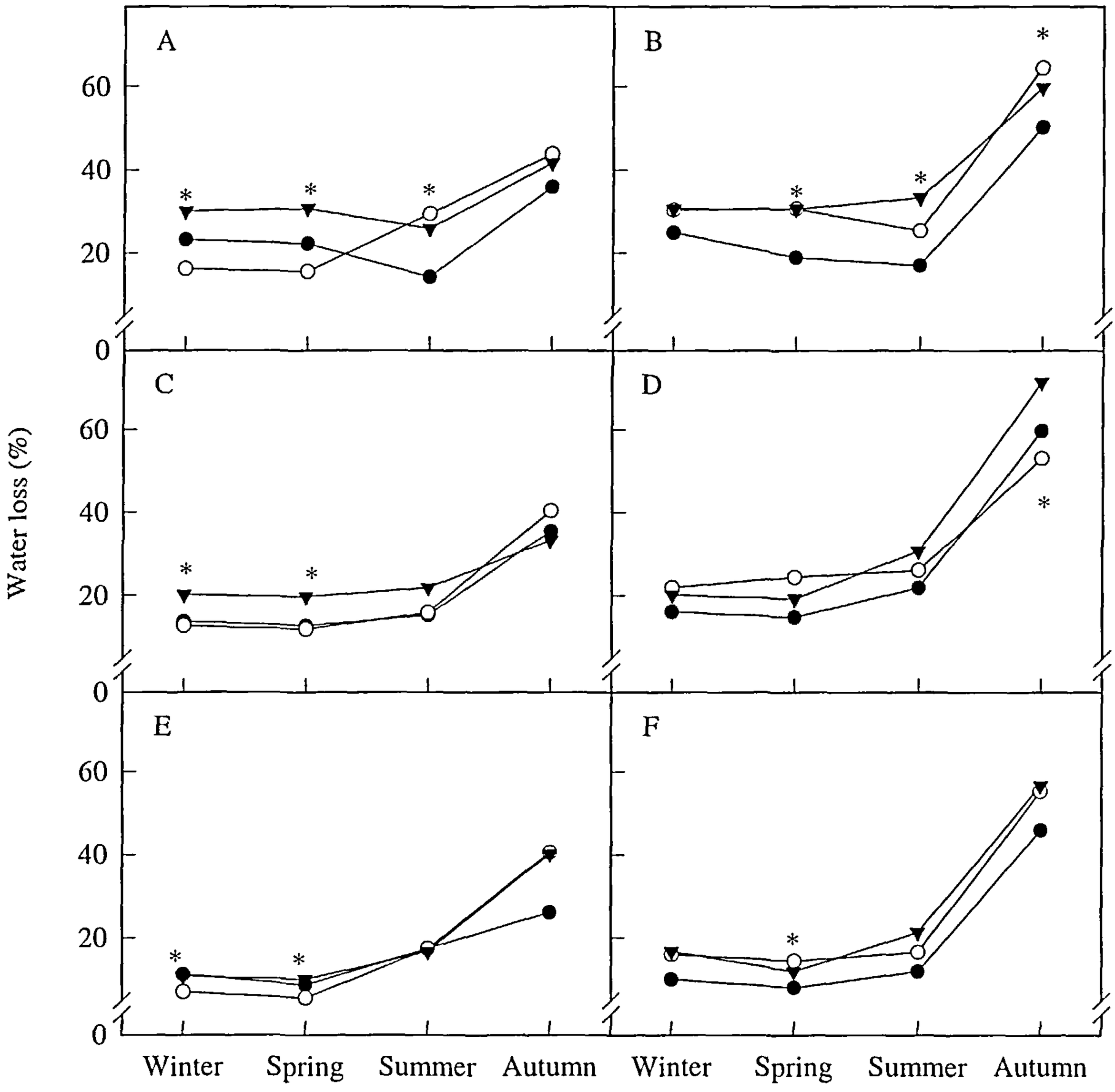


Figure 4.12: Changes in water loss (%) of detached leaves from 'First Red' (A, C, E) and 'Akito' (B, D, F) roses grown during four seasons and then stored at 1 (●), 5 (○) and 10°C (▼) and 75 (A, B), 85 (C, D) and 95% (E, F) RH. Data are means of water loss measured 12, 24 and 48 h after detachment; n = 10. Stars indicate significant difference between storage temperatures for each RH level and season at P = 0.05 (Appendix 4.2.1, Tables A4.2.1.2 – A4.2.1.25).

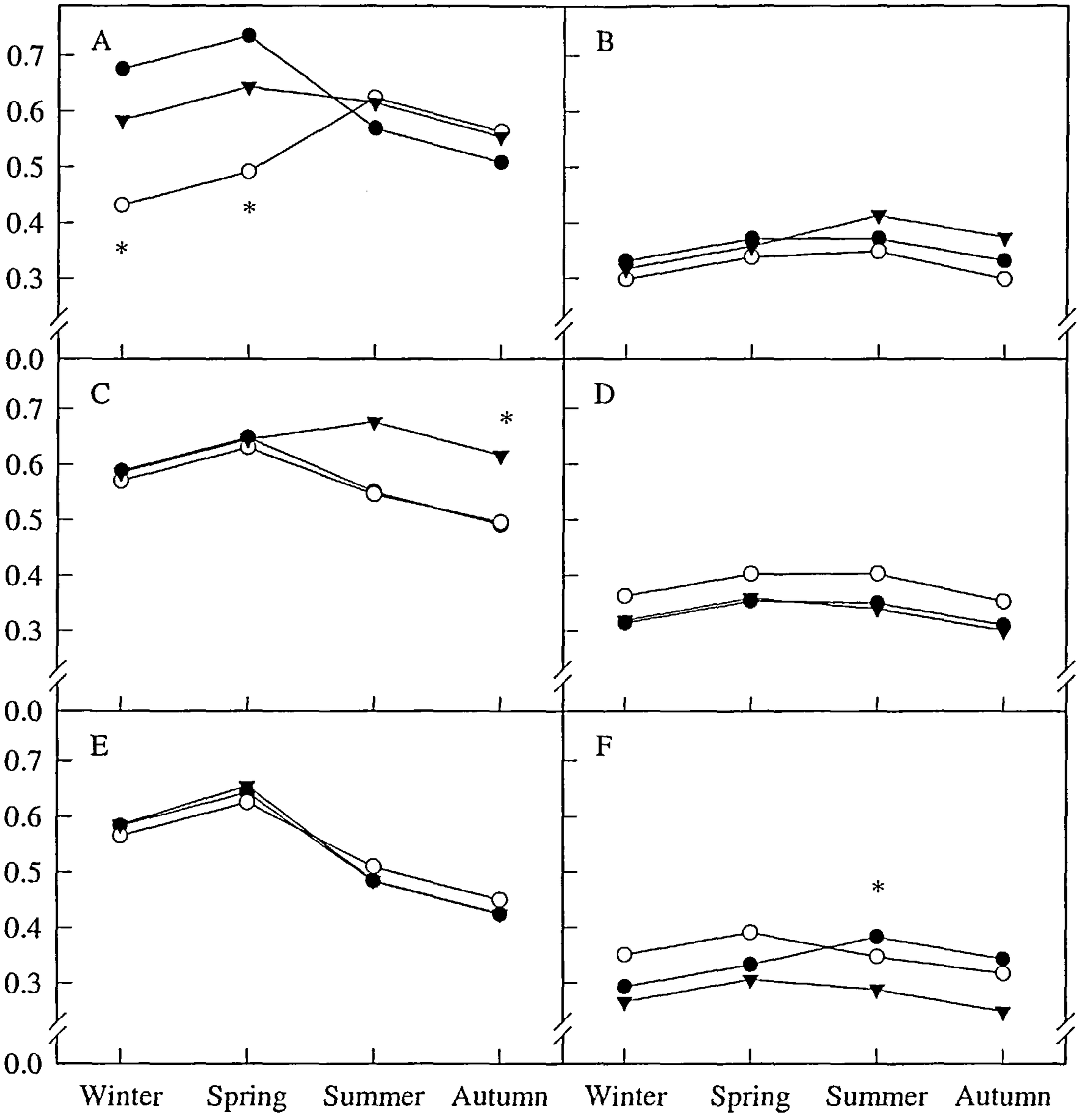


Figure 4.13: Changes in dry weight (g) of detached leaves from 'First Red' (A, C, E) and 'Akito' (B, D, F) roses grown during four seasons and then stored at 1 (●), 5 (○) and 10°C (▼) and 75 (A, B), 85 (C, D) and 95% (E, F) RH. Data are means of water loss measured 12, 24 and 48 h after detachment; n = 10. Stars indicate significant difference between storage temperatures for each RH level and season at P = 0.05 (Appendix 4.2.2, Tables A4.2.2.2 – A4.2.2.25).

4.2.2 Vase life parameters in relation to pre-harvest environmental conditions

4.2.2.1 Glasshouse environmental conditions

During winter months from December to February, PFD reached the minimum of about 780 μmol m^{-2} s^{-1} (Figure 4.14C). Mean day and night minimum temperatures were 20 and 15°C, respectively (Figure 4.14A). As a consequence of low temperature, RH reached a maximum during winter months (Figure 4.14B). In spring and summer months (from March to August), however, mean day and night temperatures and PFD sharply increased reaching values of >25, >16°C and >1200 μmol m^{-2} s^{-1}, respectively.

4.2.2.2 Correlative effects of environmental conditions on vase life parameters

In 'Akito' roses, increasing PFD during the year was significantly ($P \leq 0.05$) correlated with increased vase life, ($r^2 = 0.96$), F_v/F_m ($r^2 = 0.85$) and flower stage ($r^2 = 0.97$) (Table 4.10). Similarly, increasing PFD was linearly correlated with vase life ($P \leq 0.01$, $r^2 = 0.99$) and F_v/F_m ($P \leq 0.05$, $r^2 = 0.81$) of 'First Red' roses. Significant ($P \leq 0.05$) linear correlations between growing temperature and length of vase lives were also found for 'First Red' ($r^2 = 0.83$) and 'Akito' ($r^2 = 0.97$) roses. Moreover, the vase life of 'Akito' roses was negatively correlated ($P \leq 0.05$, $r^2 = -0.88$) with RH. The negative correlative effect of RH on vase life is presumably a consequence of the interdependence of temperature and RH; i.e. inverse relationship (Figure 4.14). Finally, solution turbidity of 'First Red' roses was strongly correlated ($P \leq 0.01$, $r^2 = 0.99$) with growing temperature during the year.

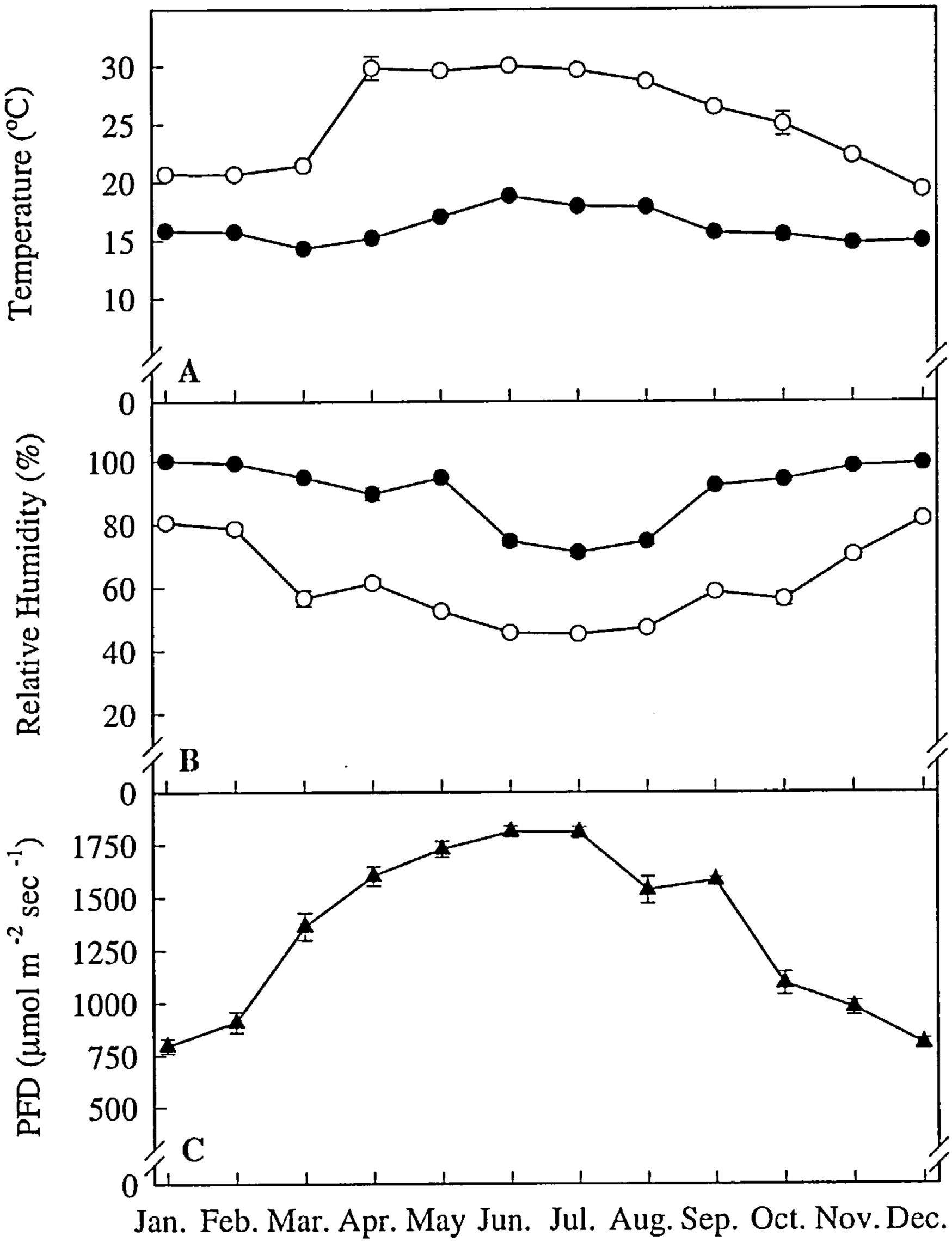


Figure 4.14: Changes in (A) temperatures (°C) and (B) RH (%) during the day (○) and night (●), and (C) Photon Flux Density (PFD) (µmol m^{-2} s^{-1}) in the glasshouse environments throughout the year. Data are means of 12 recordings per day; *n* = number of days each month. Vertical bars show ± SE for each month (*n* = number of days each month).

Table 4.10: Summary of regression (r^2-values) matrices for linear associations (y = a.x +/- b) between growing conditions (RH, temperature and PFD) in the greenhouses and vase life, F_v/F_m on d 0, flower stage on d 8, absorbance and solution turbidity for 'First Red' and 'Akito' roses.

Vase life variables	Growing conditions		
	Temperature (°C)	RH (%)	PFD (µmol m^{-2} s^{-1})
'First Red'			
Vase life	0.83 *	-0.69 n.s.	0.99 **
F_v/F_m (day 0)	0.73 n.s.	-0.80 n.s.	0.81 *
Flower stage (day 8)	-0.13 n.s.	0.11 n.s.	0.35 n.s.
Solution turbidity	0.99 **	0.95 n.s.	0.91 n.s.
'Akito'			
Vase life	0.97 *	-0.88 *	0.96 *
F_v/F_m (day 0)	0.75 n.s.	-0.80 n.s.	0.85 *
Flower stage (day 8)	0.72 n.s.	-0.60 n.s.	0.97 *
Solution turbidity	0.26 n.s.	-0.05 n.s.	0.67 n.s.

y-variables = row headings, x-variables = column headings; * $P \leq 0.05$; ** $P \leq 0.01$; n.s., not significant.

4.3 DISCUSSION

Although 'Akito' roses were more sensitive to bent neck disorder, they had generally longer vase life than 'First Red' roses. In most cases, the increased incidence of bent neck for 'Akito' roses was recorded at the end of vase life and, thus, this disorder did not markedly affect vase life duration. The greater sensitivity of 'Akito' roses to bent neck was possibly related to water deficit stress during vase life and/or a relative lack of lignification in the peduncle region (Zieslin *et al.*, 1978; van Doorn, 1997). This increased sensitivity of 'Akito' roses to bent neck is further discussed and investigated using histological techniques in Chapter 5.

4.3.1 Season effects on vase life parameters

Significant vase life and fresh weight losses were recorded for 'First Red' and 'Akito' roses grown in winter. Longer vase life for both cultivars was linearly correlated with increasing growing temperature and PFD during the year. These associations may be attributed to pre-harvest PFD levels and/or temperature effects on photosynthetic activity (Ueda *et al.*, 2000) and, as a result, carbohydrate status (Zieslin and Mor, 1990). Similarly, Slootweg *et al.* (2001) reported that vase life of summer-grown roses, with high carbohydrate levels and increased water absorption after harvest, was greater compared to autumn- and winter-grown roses. A negative correlative relationship between RH and vase life duration was also found for 'Akito' roses. High RH-associated alterations in stomatal morphology have been suggested to induce excessive water loss by flower stems (Torre and Fjeld, 2001). Such an effect may be a contributing factor to the shorter vase life of autumn- and winter-harvested roses. This explanation is also supported in autumn by the results of excessive water loss by detached leaves and increased solution usage by flower stems. However, increased water loss and solution usage was not confirmed in winter experiments as previously reported by Torre and Fjeld (2001) for roses grown in high-RH conditions. Moreover, there was no correlation between growing conditions and water loss or solution usage. Thus, the loss of vase life in winter is not attributed to water loss or solution usage by rose stems but is apparently due to low photosynthesis during cultivation, which resulted in lower carbohydrate pools as discussed above. The reduced photosynthetic activity, which possibly resulted in lower concentration of

organic compounds (Slootweg *et al.*, 2001), may also explain the lower dry weight of detached leaves recorded at harvest in autumn and winter months.

The maximum potential quantum yield of PS II (dark adapted F_v/F_m) on day 0 of vase life averaged over growing season and storage temperatures was greater for 'First Red' than for 'Akito' roses. Both cultivars had significantly lower F_v/F_m when harvested in winter. The lower F_v/F_m during winter was due to reduction in F_v/F_m of stored flowers, while F_v/F_m of non-stored control roses remained close to the maximum value of 0.83 all year round. Thus, the reduction in F_v/F_m during winter is not due to direct effects of growing season but is due to an interaction of growing season and cold storage. This consistent finding, not previously reported for roses, suggests increased sensitivity of winter-grown roses to LTI. As PFD was significantly ($P \leq 0.05$) correlated with F_v/F_m, the decline in F_v/F_m of winter-grown roses suggests that low light promotes low temperature sensitivity. Low light-induced sensitivity of plant tissues to CI through reduced accumulation of photosynthetic products has been established. Reduced carbohydrate status under low-light or dark conditions has been associated with increased chilling sensitivity of tomato leaves (King *et al.*, 1988). Moreover, provision of exogenous soluble carbohydrate can ameliorate CI (Raison and Lyons 1986) in susceptible tomato leaf tissue.

Growing roses in autumn and winter months reduced degree of flower opening. In 'Akito' roses, flower stage of roses on d 8 of vase life was positively correlated with PFD changes during the year, indicating a strong association between flower opening and light intensity in glasshouse. Marissen (2001) reported that the levels of lighting in glasshouse clearly influence the level of carbohydrates in leaves and flower buds and, as a result, flower opening. The lack of adequate starch and/or carbohydrate pool in the corolla of roses under low light conditions (e.g. autumn and winter may be cause of poor opening during vase life (van Doorn *et al.*, 1991b; Kuiper *et al.*, 1996). Carbohydrates may provide ATP for active transport of substances into the expanding corolla tissue (van Doorn *et al.*, 1991b) and possibly contribute to the decrease in osmotic potential, which drives cell elongation (Ho and Nichols 1977; van Doorn *et al.*, 1991b).

Petal colour visually changed from its original red colour to blue shade (represented by decreased b^* values) for 'First Red' roses grown from autumn to winter. This change in petal colour of roses grown in low light intensities (i.e. PFD)

of autumn and winter months may be due to inhibition of anthocyanin synthesis (Chalker-Scott, 1999). It has been reported that anthocyanins are photo-induced and may have a photo-protective function, either against light-induced photo-oxidation or against UV-B damage (Drumm-Herrel, 1984; Chalker-Scott, 1999). Thus, the possible low anthocyanin concentration in petals during autumn and winter months may have an effect on petal blueing development after harvest. b^* values decreased progressively for 'First Red' roses during vase life, indicating the development of blueing. This progressive increase in petal blueing has been attributed to pH increase occurring in the micro-environment of the anthocyanins, probably in the cell valuoles (Borochov *et al.*, 1976a; Oren-Shamir *et al.*, 2001). This pH increase occurs even though there is no change either in the pigment concentration per petal or in the ratio between the two anthocyanidins, cyanidin and pelargonidin (Biolley and Jay, 1993). In cut 'Mercedes' roses, increased cell sap pH of petals was strongly correlated (r^2 = 0.85) with b^* value decline (e.g. petal blueing) during vase life (Oren-Shamir *et al.*, 2001).

Growing roses in spring and summer enhanced vase solution turbidity and absorbance at the end of vase life. This significant effect of growing season was marked for both cultivars. Additionally, solution turbidity changes of 'First Red' roses at the end of vase life were strongly correlated ($P \leq 0.01$, $r^2 = 0.99$) with changes of growing temperature. Increased growing temperature in spring and summer may increase microbial load in rose stems during cultivation. These microbes and their degradation products can be found in vase water after harvest to increase turbidity (van Doorn, 1997; Knee, 2000). Moreover, higher growing temperatures during summer months have been found to promote flower respiration after harvest and, as a result, rose stem senescence with increased membrane breakdown and/or ion leakage (Celikel and Karacaly, 1995; Shin *et al.*, 2001). The increased water stress at higher temperature may also enhance senescence-associated changes (e.g. increased membrane breakdown and/or ion leakage) by increasing the levels of senescence hormone (e.g. ABA and ethylene) (Apelbaum and Yang, 1981). In these cases, metabolites such as carbohydrates, amino acids and amides (Halevy *et al.*, 1974) could leach into the vase solution to become substrates for microbial growth (Pompodakis *et al.*, 2004).

4.3.2 Storage effects on vase life parameters during the year

Storage of 'First Red' and 'Akito' roses for 10 days significantly ($P < 0.01$) reduced vase life compared to non-stored control flowers throughout the year. As vase life was significantly correlated with fresh weight, the reduced vase life of stored roses was associated with less capacity of roses in maintaining fresh weight. The reduction in vase life after storage was characterised by advanced wilting of leaves and fading of petals. Greater vase life losses were associated with increasing storage temperature from 1 to 10°C. Thus, vase life of 'First Red' and 'Akito' roses averaged over the growing seasons was shortest after storage at 10°C. In the case of 'First Red', the reduced vase life at 10°C compared to 1 and 5°C was consistent all year round. In 'Akito' roses, the shortest vase life at 10°C was recorded only during summer, while no significant ($P > 0.05$) differences were determined in vase life between storage temperatures from autumn to spring. Early termination of vase life after storage at 10°C was evidently due to enhanced senescence at this relatively higher storage temperature. Senescence in roses involves a sequence of events in which a rise in ethylene production precedes the increase in membrane permeability (Faragher and Mayak, 1984). As a result of greater membrane permeability, ion leakage increases leading to the visible end of vase life (Faragher *at al.*, 1986; Rubinstein, 2000).

Non-stored roses had the highest F_v/F_m. F_v/F_m for roses stored at 1°C was significantly ($P<0.01$) lower than F_v/F_m values of non-stored control roses and roses stored at 5 and 10°C. However, this effect was only observed during winter experiments when PFD and temperatures were low and, as discussed above, was attributed to low light-induced sensitivity of plant tissues to CI. A similar reduction in F_v/F_m has also been reported for kangaroo paw flowers after storage at 0°C (Joyce and Shorter, 2000; Miranda *et al.*, 2000). In contrast to roses, the fall of F_v/F_m in kangaroo paw was recorded for flowers grown between spring and summer and was significantly correlated ($r^2 = 0.83$, $P \leq 0.05$) with vase life decline. However, in the current experiments, although there was significant correlation ($P \leq 0.01$) between F_v/F_m on day 0 and vase life, this relationship was very weak; $r^2 = 0.43$ and 0.44 for 'First Red' and 'Akito' roses, respectively. Thus, the acute decline in F_v/F_m after storage at 1°C did not translate into a correspondingly sharp vase life reduction

(Pompodakis *et al.*, 2005). The fall in F_v/F_m value for 'Akito' roses on day 0 of vase life was strongly correlated ($r^2 = 0.97$) with reduced storage temperature. Significant and almost linear reduction in F_v/F_m ratio may be attributed to progressively worsening physicochemical lesions related to LTI or CI that affect the energy transfer pathway from antennae to reaction centres (Krause and Weis, 1991; Miranda *et al.*, 2000). The slope of regression was less in 'First Red' roses ($r^2 = 0.67$) than in 'Akito' roses ($r^2 = 0.97$). Thus, 'First red' was less sensitive to LTI than 'Akito'.

Storage of roses at low temperature significantly ($P < 0.05$) reduced flower opening. This adverse effect of storage on flower opening was consistently recorded for both 'First Red' and 'Akito' roses throughout the year. Storing roses at 1°C resulted in minimum flower opening. As flower opening in roses is an integrated result of petal expansion and reflexing (Mayak *et al.*, 2001), low temperature-induced alterations in these two processes (e.g. enhanced water stress) could have unfavourable effects. However, Faragher *et al.* (1984) showed that inhibition of flower opening at low temperature cannot be explained in terms of water loss but is attributable to increased ethylene production. Indeed, increase in 1-aminocyclopropane-1-carboxylic acid (ACC) and malonyl-ACC (MACC) levels in the petals of 'Visa' roses during cold storage indicated that the ACC-synthetase enzyme remains active enhancing ethylene biosynthesis (Serrano *et al.*, 1992). Ethylene could inhibit petal reflexing through its effects on membrane permeability, which involves changes in protein and enzyme synthesis (Faragher and Mayak, 1984).

Moreover, the significant increase in petal blueing (decreased b^* value) of stored roses reported here is apparently due to continued senescence during cold storage. Faragher and Mayak (1984) supported that the increased membrane permeability and the subsequent ion leakage during cold storage were primary events of petal blueing. However, recently, petal blueing has been greatly associated with pH increase in petals during senescence rather than ion leakage (Oren-Shamir *et al.*, 2001). Accordingly, petal blueing is the result of biochemical processes (e.g. increase in hydrolytic enzymes and a drop in protein levels) that can increase the vacuolar pH due to ammonia release. Finally, stored roses had more advance foliage stages during vase life, indicating premature wilting of foliage. Increased foliage stages were significantly ($P \leq 0.01$) correlated with vase life decline in 'First Red' ($r^2 = -0.48$) and 'Akito' roses ($r^2 = -0.35$), respectively. An increase in ion leakage, reflecting

increased membrane permeability occurs before the visible symptoms of wilting (Faragher and Mayak, 1984). These events may be due to the developed LTI- and/or senescence-related changes in membrane properties.

CHAPTER 5

EFFECT OF DIFFERENT ABSCISIC ACID TREATMENTS TO IMPROVE VASE LIFE OF CUT 'FIRST RED' AND 'AKITO' ROSES STORED AT LOW TEMPERATURE

5.1 INTRODUCTION

Exogenous application of ABA to chilling sensitive plants before, during or even shortly after a low temperature treatment has been shown to protect against CI or LTI. ABA helps protect plant tissues against CI by inducing stomatal closure (Janowiak *et al.*, 2002; Pardossi *et al.*, 1992), enhancing antioxidant enzyme activity (Prasad *et al.*, 1994) and by modulating free polyamine levels (Lee *et al.*, 1997). ABA induced chilling tolerance in *Arabidopsis thaliana* by enhancing the expression of *ABI3* gene (Pearce, 1999; Tamminen *et al.*, 2001).

When chilling sensitive plants, such as *Lycopersicon esculentum* L. (Starck *et al.*, 1998), *Zea mays* L. (Janowiak and Dorffling, 1996; Janowiak *et al.*, 2002; Aroca *et al.*, 2003), *Oryza sativa* L. (Lee *et al.*, 1997), *Phaseolus vulgaris* L. (Pardossi *et al.*, 1992) and *Euphorbia pulcherrima* (Tantau *et al.*, 1991), are exposed to chilling temperatures, their endogenous ABA level increases. For example, in maize leaves that were chilled for two days at 4°C, the ABA content was generally higher at the end of chilling period, especially in the tolerant genotypes. Transfer of the chilled leaves to warmer conditions resulted in a decrease of the ABA content within two days to nearly the same level as that before chilling (Janowiak *et al.*, 2002). Pardossi *et al.* (1992) and Capell and Dorffling (1989) reported that in *Phaseolus vulgaris* L. and in *Cucumis sativus* L., respectively, the rise in ABA during chilling is associated with a chilling-induced water deficit. In contrast, Starck *et al.* (1998) found that in tomato the increase in ABA content is caused directly by low temperature while in maize it has been suggested that both factors (temperature and water deficit) are involved (Janowiak *et al.*, 2002). Based on their study, Capell and Dorffling (1993), Janowiak and Dorffling (1996) and Janowiak *et al.* (2002) have developed a hypothesis that the ability to produce ABA is an important prerequisite for chilling tolerance.

A positive and significant correlation ($r = 0.69$; $P \leq 0.001$) was also found

between effective quantum yield (Φ_{PSII}), which measures the efficiency of PSII photochemistry (Maxwell and Johnson, 2000), and ABA content after two days of chilling at 4°C (Janowiak *et al.*, 2002). Sensitivity of maize plants to CI measured as reduction in F_v/F_m, membrane damage and whole plant survival was increased, with reduction in endogenous ABA content (Janowiak *et al.*, 2002). Moreover, Pardossi *et al.* (1992) found a correlation (r = 0.67) between the rise in leaf ABA content and decrease in leaf conductance during chilling, indicating an effect of ABA accumulation on stomatal behaviour. However, recently Wilkinson *et al.* (2001) have found that in short-term exposure of *Commelina communis* leaves to chilling (1h), stomatal closure was due to increase in apoplastic calcium uptake by guard cells, without any change in the ABA content. During such short-term exposures to chilling, calcium ions may enter guard cells and promote stomatal closure while apoplastic calcium sensitises guard cells to ABA (Wilkinson and Davies, 2002).

This study investigated the efficacy of postharvest treatment of rose flowers with ABA in controlling LTI. Based on our previous results (Chapter 4), autumn- and winter-grown cut roses were observed to be most susceptible to LTI. Therefore, this study was focused on determining the capability of ABA to induce chilling tolerance in cut roses, which had been grown in autumn and winter months. Cut 'First Red' and 'Akito' roses were pulsed with 10^{-1} M ABA (Kohl and Rundle, 1972) or sprayed onto the leaves with 10^{-5} M ABA (Barthe *et al.*, 1991) and then stored wet at 1 and 5°C. At these concentrations, ABA has been found to be most effective in causing stomatal closure of rose, thereby resulting in lower transpiration rates (Halevy *et al.*, 1974; Borochov *et al.*, 1976). Vase life parameters (e.g. vase life duration, F_v/F_m, bent neck incidence, flower opening, fresh weight and solution usage) along with biochemical assays (e.g. electrolyte leakage, malondialdehydes and endogenous ABA concentration) were evaluated to analyse the possible role of ABA on chilling tolerance of roses. Electrolyte leakage and malondialdehyde levels were measured as indicators of membrane permeability and lipid peroxidation, respectively (Meir *et al.*, 1992; Pardossi *et al.*, 1992). Both parameters have been found to increase during senescence (Panavas *et al.*, 1998) or after CI (Murata, 1989).

5.1.1 Bent neck study

In first year results (Chapter 4, section 4.2.1.2), 'Akito' roses had great sensitivity to bent neck symptom, which is partially dependant on the degree of lignification in peduncles (van Doorn and Reid, 1995). Thus, in the present study (second year experiments), histological techniques were performed to determine the extent of lignification of vascular cells during bent neck symptom. This study was focused on measurements of lignified cells in the peduncles of 'First Red' and 'Akito' roses grown from autumn 2003 to winter 2003-04.

5.2 RESULTS

5.2.1 Effects of storage temperature and ABA treatments on vase life of roses grown from autumn 2003 to winter 2003 – 04.

5.2.1.1 Vase life

Storage of 'First Red' roses for 10 days significantly ($P < 0.05$) shortened flower and foliage lives (Table 5.1, Appendix 5.1.1 and 5.1.2, Tables A5.1.1.1, A5.1.1.2, A5.1.2.1 and A5.1.2.2). Shortest vase lives for 'First Red' roses were recorded after storage at 5°C for 10 days. In 'Akito' roses, although flower life did not differ markedly between storage treatments (Table 5.1, Appendix 5.1.1, Table A5.1.1.3), foliage life was significantly ($P < 0.05$) reduced at 1°C (Appendix 5.1.1, Table A5.1.2.3). ABA treatment did not affect significantly vase life of 'First Red' roses (Table 5.1, Figure 5.1A, C). However, pulsing 'Akito' roses for 24 hours with 10^{-1} M ABA extended both flower and foliage lives (Table 5.1).

Table 5.1: Effects of storage and ABA treatments on flower and foliage lives of 'First Red' and 'Akito' roses. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were pulsed or sprayed with ABA or put into vases without ABA (control). Data are main-factor $\bar{x}$; n = 36. Data for independent treatment means are presented in Figure 5.1. ANOVA and Duncan's tests are presented in Appendix 5.1.

Factors	**'First Red'**		**'Akito'**	
	Flower life [a]	**Foliage life**	**Flower life**	**Foliage life**
1) Storage treatment [b]				
control	10.2a	13.2a	12.2a	12.1a
1°C	7.5b	9.6b	11.4a	7.4b
5°C	6.8b	7.7b	12.2a	11.2a
2) ABA treatment				
control	8.3a	10.0a	11.4a	9.2a
spray	8.1a	11.2a	11.0a	9.2a
pulse	8.2a	9.4a	13.4b	12.3a

[a] Data are main factor means of flower and foliage lives. [b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

ABA treatment generally increased flower and foliage lives of 'Akito' roses stored at 5ºC (Figure 5.1B, D). At this storage temperature, foliage life of 'Akito' roses pulsed with ABA was significantly different compared to controls according to Duncan's multiple range test (Appendix 5.1.2, Table A5.1.2.4).

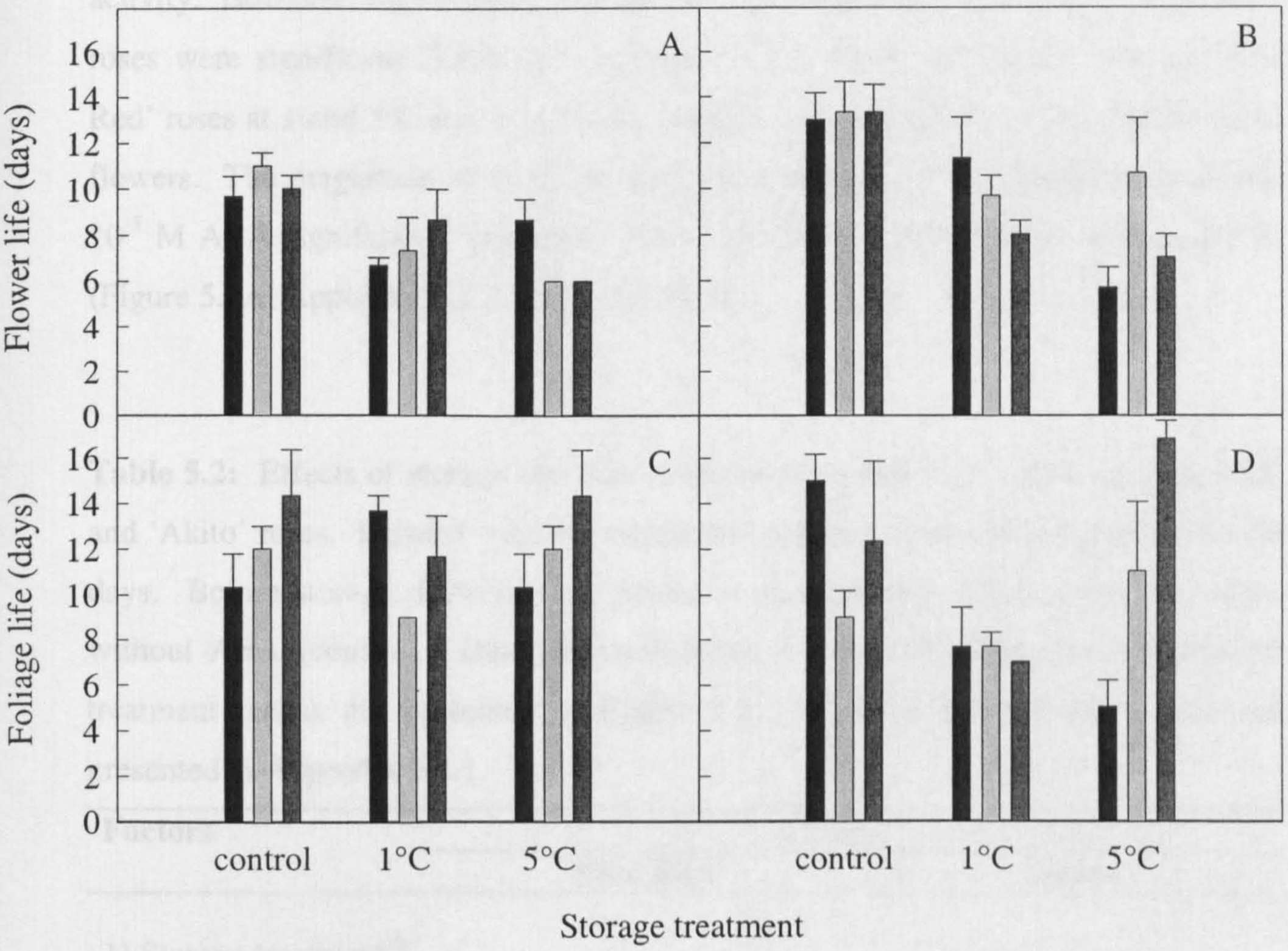


Figure 5.1: Flower and foliage lives of 'First Red' (A, C) and 'Akito' (B, D) roses grown from autumn 2003 to winter 2003 – 04 and then put into vases (control) or stored wet at 1 and 5ºC. Flowers were put into vases without ABA (■) or sprayed (■) or pulsed (■) with ABA before storage. Data are independent treatment means; n = 6. Vertical bars show ± S.E. (n = 12) for each treatment. Main factor means are presented in Table 5.1. ANOVA and Duncan's tests are presented in Appendix 5.1.

5.2.1.2 Relative chlorophyll fluorescence

F_v/F_m at the beginning of vase life (day 0)

Leaf F_v/F_m of 'Akito' roses, which was not affected by storage temperature or ABA treatment (Table 5.2, Figure 5.2B, Appendix 5.2.1, Table A5.2.1.3), remained above 0.80 after storage, indicating the lack of physicochemical damage in PSII activity. However, the effects of storage and ABA treatments on F_v/F_m of 'First Red' roses were significant (Table 5.2, Appendix 5.2.1, Table A5.2.1.1). Storing 'First Red' roses at 1 and 5°C significantly decreased F_v/F_m compared to non-stored control flowers. The magnitude of F_v/F_m decline was greatest at 1°C. Spraying roses with 10^{-5} M ABA significantly prevented F_v/F_m decline brought about by storage at 1°C (Figure 5.2A, Appendix 5.2.1, Table A5.2.1.2).

Table 5.2: Effects of storage and ABA treatments on leaf F_v/F_m (d 0) of 'First Red' and 'Akito' roses. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were pulsed or sprayed with ABA or put into vases without ABA (control). Data are main-factor $\bar{x}$; n = 36. Data for independent treatment means are presented in Figure 5.2. ANOVA and Duncan's tests are presented in Appendix 5.2.1.

Factors	F_v/F_m [a]	
	'First Red'	**'Akito'**
1) Storage treatment [b]		
control	0.830a	0.833a
1°C	0.696b	0.810a
5°C	0.778c	0.818a
2) ABA treatment		
control	0.761a, b	0.811a
spray	0.796a	0.809a
pulse	0.761b	0.805a

[a] Data are main factor means of F_v/F_m on d 0. [b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

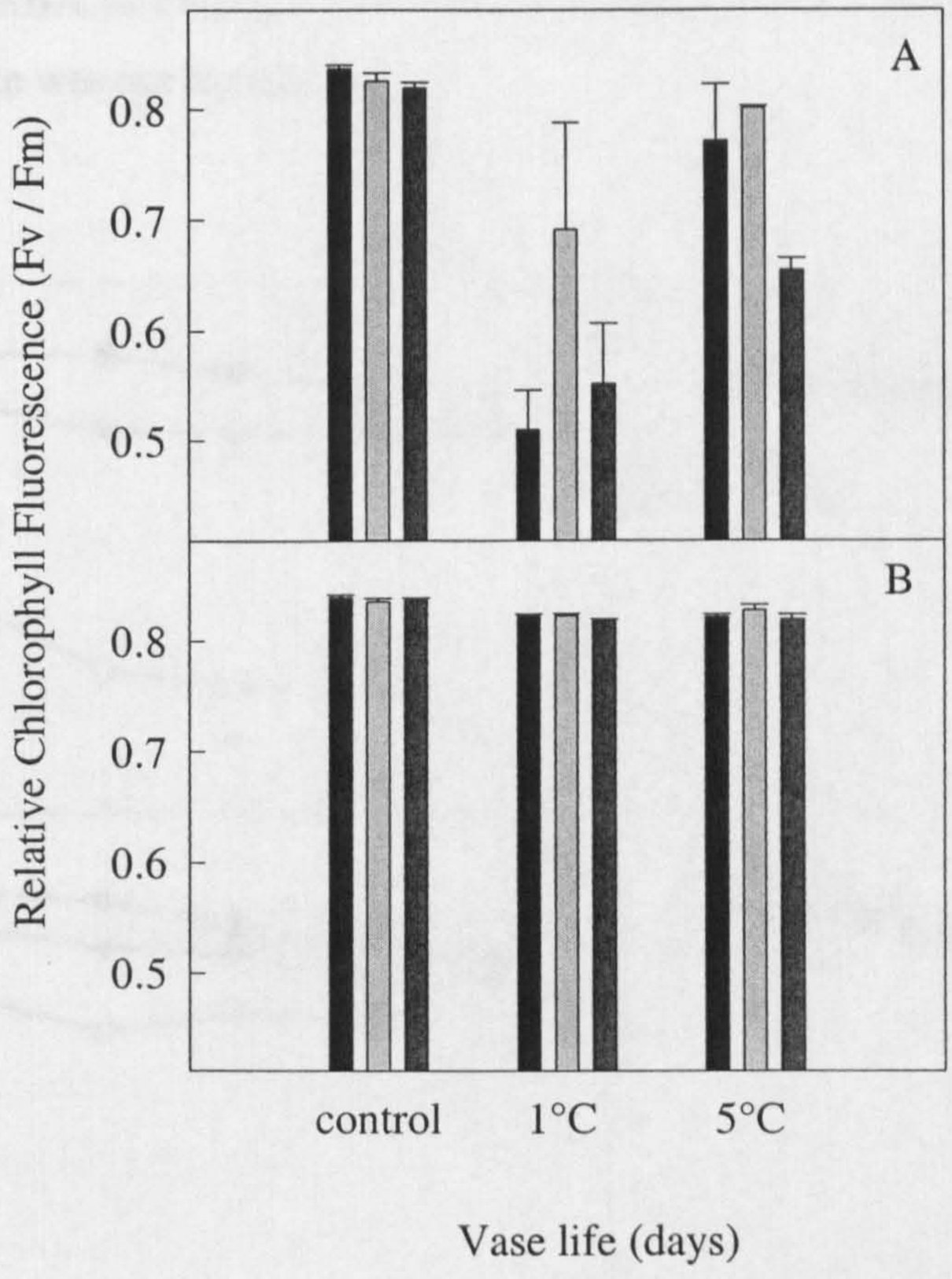


Figure 5.2: Leaf F_v/F_m on d 0 of 'First Red' (A) and 'Akito' (B) roses grown from autumn 2003 to winter 2003 – 04 and then put into vases (control) or stored wet at 1 and 5°C. Flowers were put into vases without ABA (■) or sprayed (■) or pulsed (■) with ABA before storage. Data are independent treatment means; n = 6. Vertical bars show ± S.E. (n = 12) for each treatment. Main factor means are presented in Table 5.2. ANOVA and Duncan's tests are presented in Appendix 5.2.1.

F_v/F_m changes during vase life

Non-stored 'First Red' flowers had comparatively less F_v/F_m losses during vase life than those stored at 1 and 5°C (Figure 5.3A, C, E, Appendix 5.2.2, Tables A5.2.2.1 – A5.2.2.15). 'First Red' roses had largest losses in F_v/F_m value after storage at 1°C. The reduced F_v/F_m at 1°C was significantly different on days 0 and 8 in control and days 0 and 4 in pulse treatments, respectively. F_v/F_m of 'Akito' roses did not differ significantly between storage and ABA treatments during vase life (Figure 5.3B, D, F, Appendix 5.2.2, Tables A5.2.2.16 – A5.2.2.30). In 'Akito' roses

pulsed with ABA, storage at 1°C resulted in larger F_v/F_m losses after day 4; however, this difference was not significant.

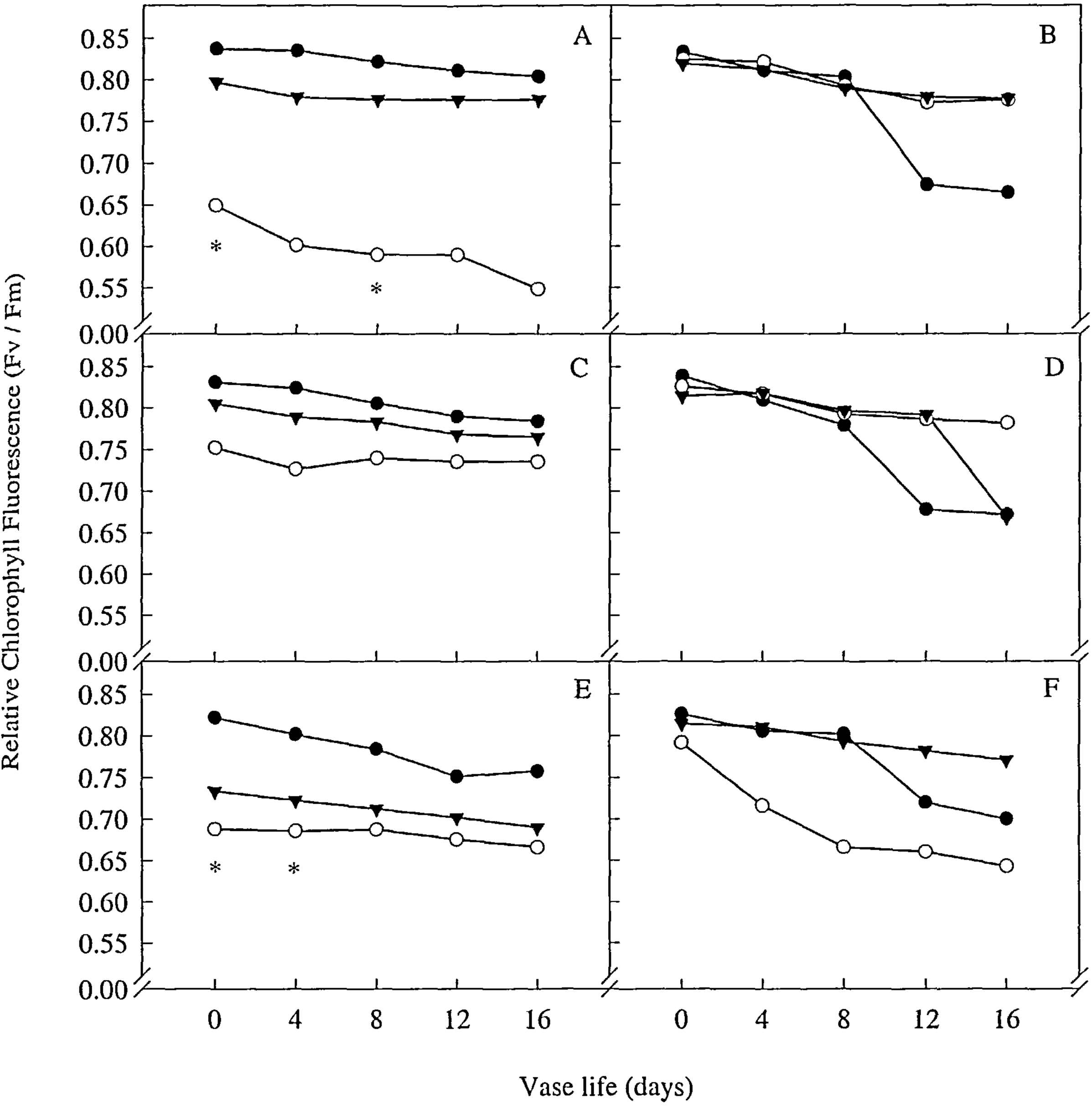


Figure 5.3: F_v/F_m changes during vase life for 'First Red' (A, C, E) and 'Akito' (B, D, F) roses grown from autumn 2003 to winter 2003 – 04 and then put into vases (control) (●) or stored wet at 1 (○) and 5°C (▼). Before storage, flowers were sprayed (C, D) or pulsed (E, F) with ABA or put into vases without ABA (A, B). Data are independent treatment means; n = 12. Stars indicate significant differences between storage treatments for each day at P = 0.05 (Appendix 5.2.2).

5.2.1.3 Flower condition

Neither storage nor ABA treatments affected bent neck incidence of roses (Table 5.3, Appendix 5.3.1, Tables A5.3.1.1 – A5.3.1.2). 'Akito' roses had comparatively larger bent neck incidence than 'First Red' roses. Increased sensitivity of 'Akito' roses to bent neck was consistently recorded for both control and stored roses either with or without ABA (Table 5.3). Furthermore, bent neck symptoms were recorded earlier for 'Akito' than for 'First Red' roses during vase life evaluation (personal observations). The early development of bent neck symptoms markedly reduced flower life of 'Akito' roses. Storing 'First Red' roses at 1 and 5°C generally resulted in less maintenance of corolla diameter during vase life (Figure 5.4A, C, E, Appendix 5.3.2). In 'Akito' roses, there were no significant differences in corolla diameter between treatments (Figure 5.4B, D, F, Appendix 5.3.2).

Table 5.3: Bent neck incidence of 'First Red' and 'Akito' roses stored wet at 1 and 5°C or were not stored (control). Before storage, flowers were sprayed or pulsed with ABA or put into vases without ABA (control). Individual treatment data are $\bar{x}$; n = 12. ANOVA tables are presented in Appendix 5.3.1.

Storage treatment	ABA treatment	Bent neck (%) [a]	
		'First Red'	'Akito'
Control	control	0.0a	66.6a
	spray	16.6a	33.3a
	pulse	16.6a	33.3a
1°C	control	16.6a	33.3a
	spray	16.6a	33.3a
	pulse	33.3a	66.6a
5°C	control	16.6a	33.3a
	spray	16.6a	33.3a
	pulse	0.0a	0.0a
10°C	control	14.8a	66.6a
	spray	0.0a	33.3a
	pulse	16.6a	33.3a
Column means		13.7	38.8

[a] Within columns, numbers followed by the same letter are not significantly different at P = 0.05.

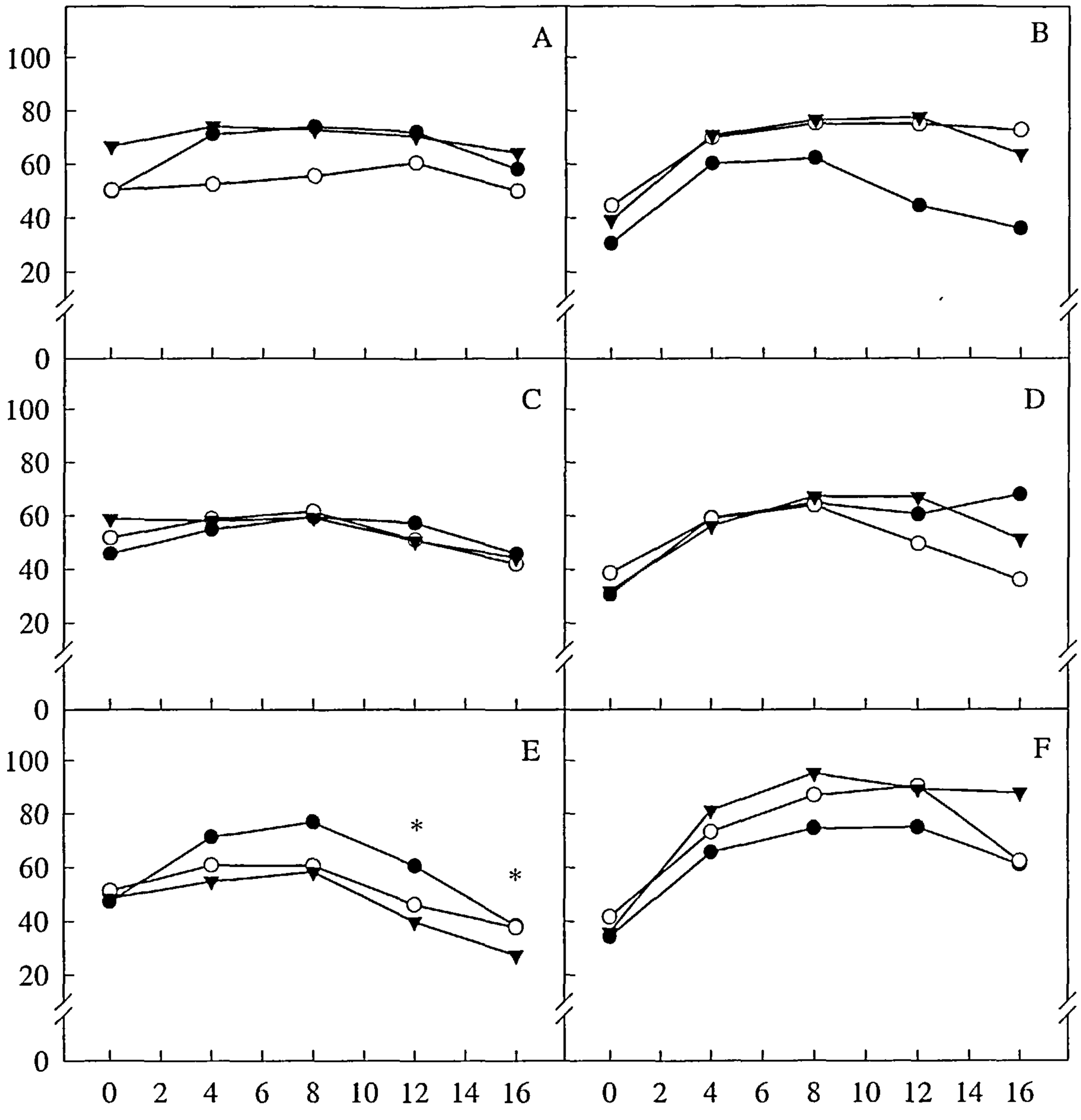


Figure 5.4: Changes in corolla diameter during vase life for 'First Red' (A, C, E) and 'Akito' (B, D, F) roses grown from autumn 2003 to winter 2003 – 04 and then put into vases (control) (●) or stored wet at 1 (○) and 5°C (▼). Before storage, flowers were sprayed (C, D) or pulsed (E, F) with ABA or put into vases without ABA (A, B). Data are independent treatment means; n = 12. Stars indicate significant differences between storage treatments for each day at P = 0.05 (Appendix 5.3.2).

5.2.1.4 Relative fresh weight and solution usage

Fresh weight of 'Akito' roses was better maintained than that of 'First Red' throughout vase life (Figure 5.5). Fresh weight of 'First Red' fell below the initial weight of 100% after day 2 and, thereafter, progressively decreased until the end of vase life. Conversely, 'Akito' roses gained fresh weight above the initial 100% until day 6. Although, there was no further increase in fresh weight of 'Akito' roses after day 6, fresh weight of 'Akito' was maintained greater than that of 'First Red' until the end of vase life. In 'First Red' roses, non-stored flowers had significantly ($P < 0.05$) less fresh weight loss during vase life evaluation than flowers stored at 1 and 5°C for any of the ABA treatments (Figure 5.5A, C, E, Appendix 5.4.1, Tables A5.4.1.1 – A5.4.1.24). In control and pulse ABA treatments, 'First Red' had greater capacity in maintaining fresh weight when stored at 1°C rather than at 5°C (Table 5.5A, E, Appendix 5.4.1, Tables A5.4.1.1 – A5.4.1.8 and A5.4.1.17 – A5.4.1.24). Stored 'Akito' had significantly ($P < 0.05$) greater fresh weight losses than non-stored 'Akito' roses on day 2 either with or without ABA (Figure 5.5B, D, F, Appendix 5.4.1, Tables A5.4.1.25, A5.4.1.33 and A5.4.1.41). However, after day 2, there were no significant differences in fresh weight changes between treatments for 'Akito' roses (Appendix 5.4.1, Tables A5.4.1.25 – A5.4.1.48).

Solution usage by 'Akito' roses was comparatively greater than that by 'First Red' throughout vase life for any of the ABA treatments (Figure 5.6). Solution usage by 'First Red' roses was higher during vase life evaluation when flowers were not stored. Reduced solution usage by stored 'First Red' roses was significant almost throughout first vase life (Figure 5.6A, C, E, Appendix 5.4.2, Tables A5.4.2.1 – A5.4.2.24). Storage treatments had opposite effects on solution usage by 'Akito' roses (Figure 5.6B, D, F, Appendix 5.4.2, Tables A5.4.2.25 – A5.4.2.48). For example, when roses were not treated or sprayed with ABA, storage at 1°C reduced solution usage by flowers (Figure 5.6B, D). On the other hand, when 'Akito' roses were pulsed with ABA and then stored at 5°C, they had greatest solution usage (Figure 5.6F). Vase solution was slightly reduced by 'Akito' roses, which had previously been pulsed with ABA (Figure 5.6F). However, this negligible effect of ABA was not significant.

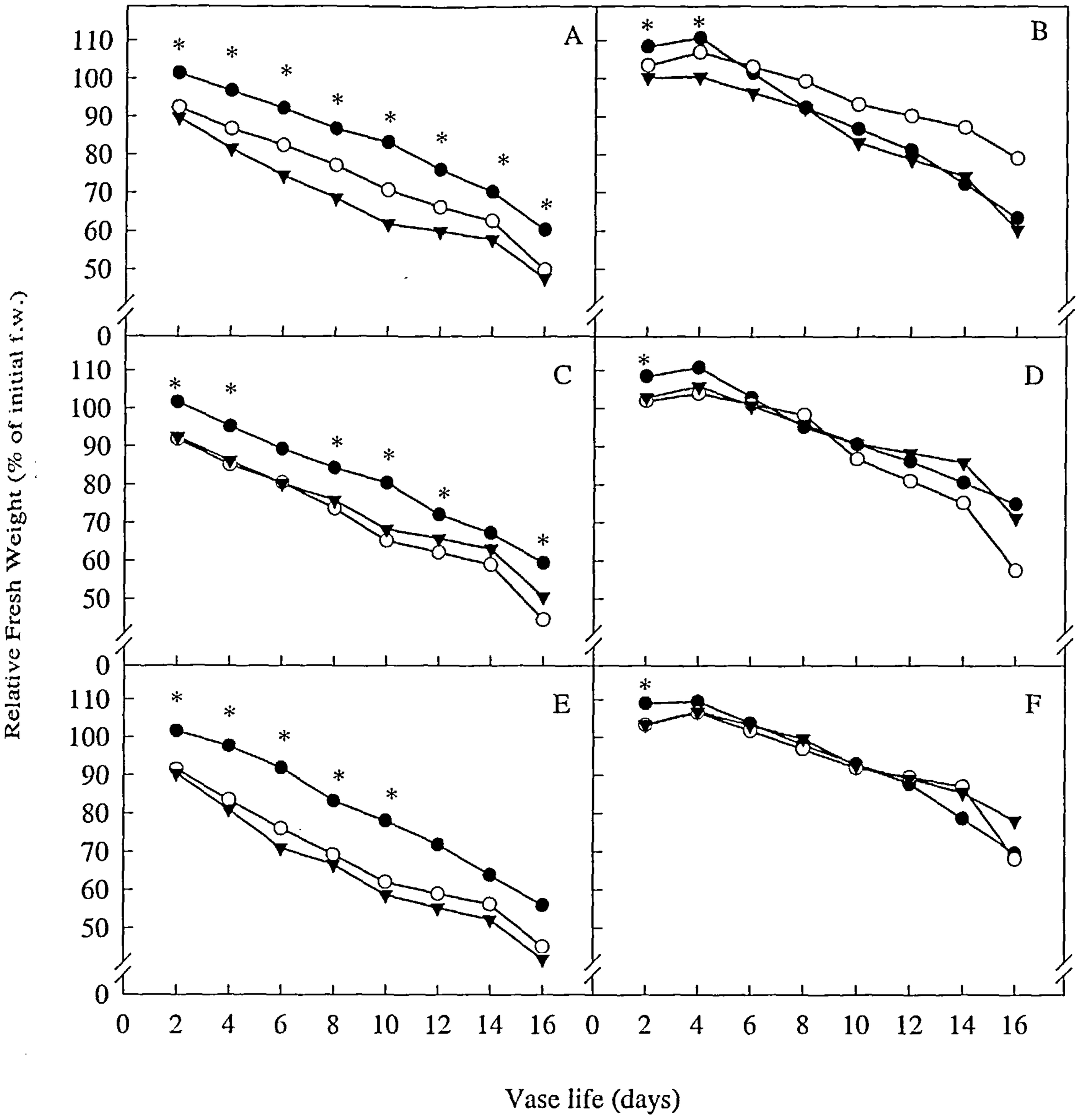


Figure 5.5: Fresh weight changes during vase life for 'First Red' (A, C, E) and 'Akito' (B, D, F) roses grown from autumn 2003 to winter 2003 – 04 and then put into vases (control) (●) or stored wet at 1 (○) and 5°C (▼). Before storage, flowers were sprayed (C, D) or pulsed (E, F) with ABA or put into vases without ABA (A, B). Data are independent treatment means; n = 12. Stars indicate significant differences between storage treatments for each day at P = 0.05 (Appendix 5.4.1).

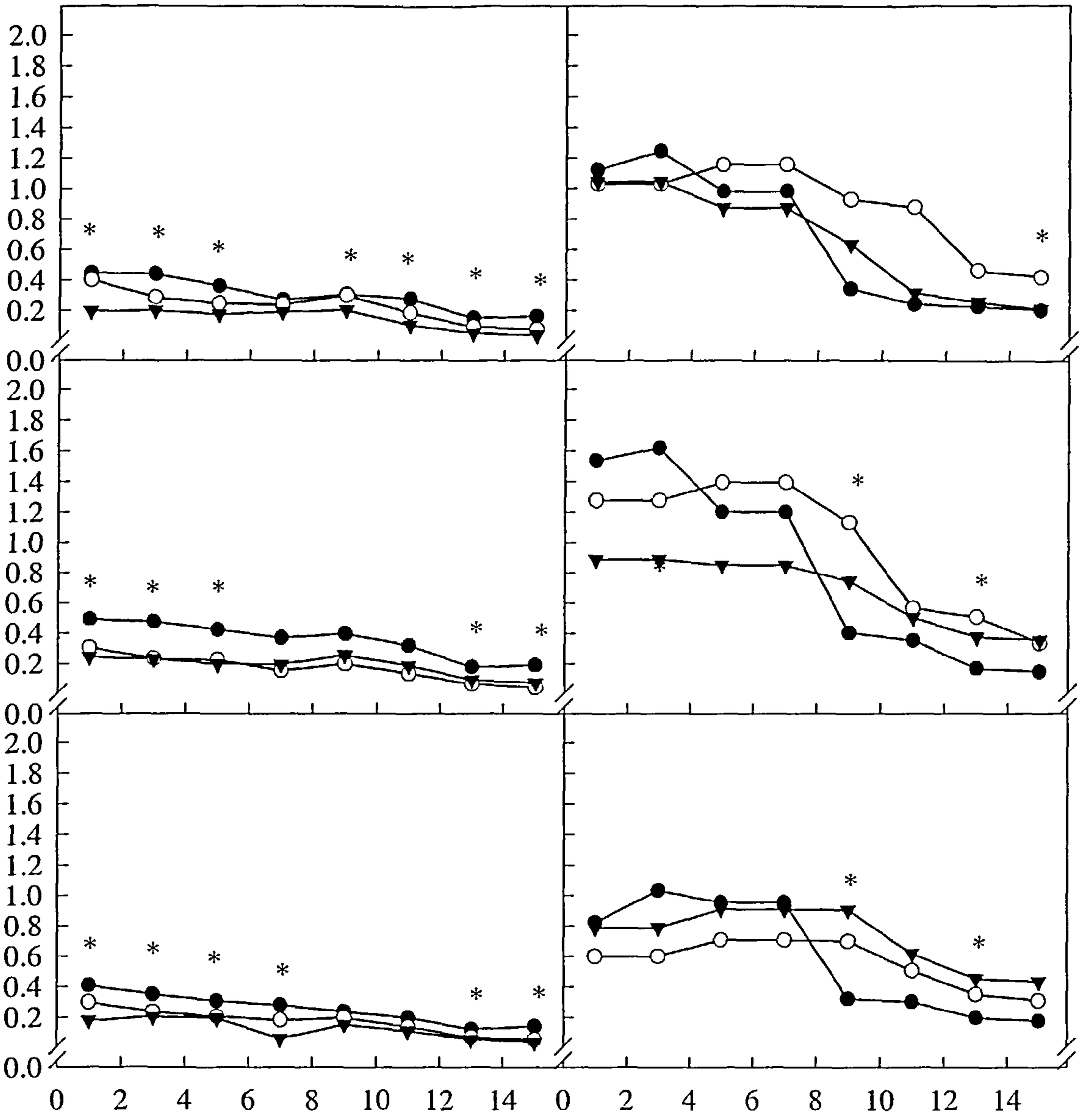


Figure 5.6: Changes in solution usage during vase life by 'First Red' (A, C, E) and 'Akito' (B, D, F) roses grown from autumn 2003 to winter 2003 – 04 and then put into vases (control) (●) or stored wet at 1 (○) and 5°C (▼). Before storage, flowers were sprayed (C, D) or pulsed (E, F) with ABA or put into vases without ABA (A, B). Data are independent treatment means; n = 12. Stars indicate significant differences between storage treatments for each day at P = 0.05 (Appendix 5.4.2).

5.2.1.5 Biochemical assays

Electrolyte leakage in petals and leaves

Electrolyte leakage in petals and leaves of roses was greater at the middle (day 10) than at the beginning (day 0) of vase life (Table 5.4). In most treatments, the increased electrolyte leakage of roses on day 10 was significantly different compared to electrolyte leakage on day 0 (Figures 5.7 and 5.8). Furthermore, in most treatments, greater electrolyte losses were recorded in leaves rather than in petals for both cultivars. Neither storage nor ABA treatments affected significantly ($P > 0.05$) electrolyte leakage in petals and leaves of 'First Red' roses (Table 5.4, Appendix 5.5.1, Tables A5.5.1.1, A5.5.1.2, A5.5.1.6 and A5.5.1.7). In 'Akito' roses, however, storing flowers for 10 days enhanced electrolyte leakage of petals and leaves (Table 5.4, Appendix 5.5.1, Tables A5.5.1.3 – A5.5.1.5 and A5.5.1.8 – A5.5.1.10). Electrolyte leakage in petals and leaves was greatest for 'Akito' roses after storage at 1°C compared to non-stored and stored roses at 5°C. This increased electrolyte leakage at 1°C was consistently recorded on days 0 and 10 (Figures 5.7 and 5.8). According to Duncan's multiple range test, when 'Akito' roses were not treated with ABA, storage at 1°C significantly increased electrolyte leakage in leaves and petals on days 0 and 10, respectively (Appendix 5.5.1, Tables A5.5.1.6 and A5.5.1.9).

ABA treatments did not affect the rate of electrolyte leakage in petals of 'Akito' roses from day 0 to 10. However, leaf electrolyte leakage was significantly reduced at the beginning of vase life (day 0), when 'Akito' roses were sprayed or pulsed with 10^{-1} and 10^{-5} M ABA, respectively. Thereafter (day 10), only pulse treatment prevented electrolyte loss in leaves of 'Akito' roses.

Table 5.4: Effects of storage and ABA treatments on electrolyte leakage (%) in petals and leaves of 'First Red' and 'Akito' roses on days 0 and 10. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were pulsed or sprayed with ABA or put into vases without ABA (control). Data are main-factor $\bar{\times}$; n = 36. Data for independent treatment means are presented in Figures 5.7 and 5.8. ANOVA and Duncan's tests are presented in Appendix 5.5.1.

Factors	'First Red'		'Akito'	
	Day 0 [a]	Day 10	Day 0	Day 10
	a. Electrolyte leakage (%) in petals			
1) Storage treatment [b]				
control	15.2a	20.1a	8.1a	14.4a
1°C	16.4a	23.3a	12.2b	32.7b
5°C	12.2a	23.7a	9.7a, b	28.0b
2) ABA treatment				
control	13.9a	21.7a	10.0a	29.8a
spray	14.3a	22.8a	10.8a	22.4a
pulse	15.5a	22.5a	9.0a	22.9a
	b. Electrolyte leakage (%) in leaves			
1) Storage treatment				
control	10.5a	17.9a	8.1a	43.7a, b
1°C	15.8a	34.5a	17.1b	54.7a
5°C	16.8a	39.7a	12.7c	24.4b
2) ABA treatment				
control	15.0a	27.3a	15.2a	56.2a
spray	10.9a	24.0a	11.1b	41.1a, b
pulse	17.2a	40.7a	11.4b	25.4b

[a] Data are main factor means of electrolyte leakage in petals and leaves. [b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

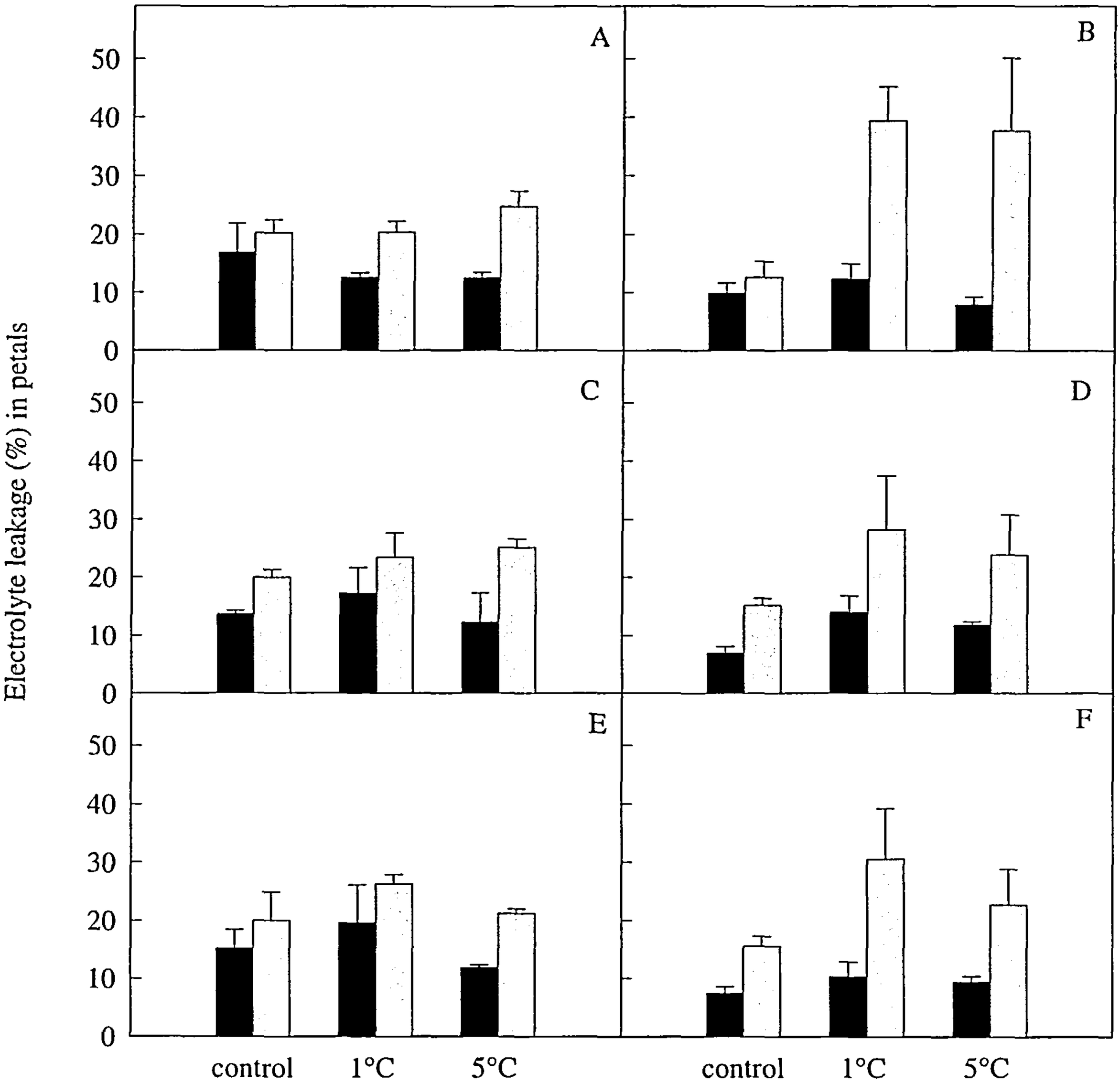


Figure 5.7: Electrolyte leakage (%) in petals of 'First Red' (A, C, E) and 'Akito' (B, D, F) roses on days 0 (■) and 10 (▣) of vase life. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were put into vases without ABA (control) (A, B) or sprayed (C, D) or pulsed (E, F) with ABA. Data are independent treatment means (± S.E.); n = 12. Main factor means are presented in Table 5.4. ANOVA and Duncan's tests are presented in Appendix 5.5.1, Tables A5.5.1.1 – A5.5.1.5.

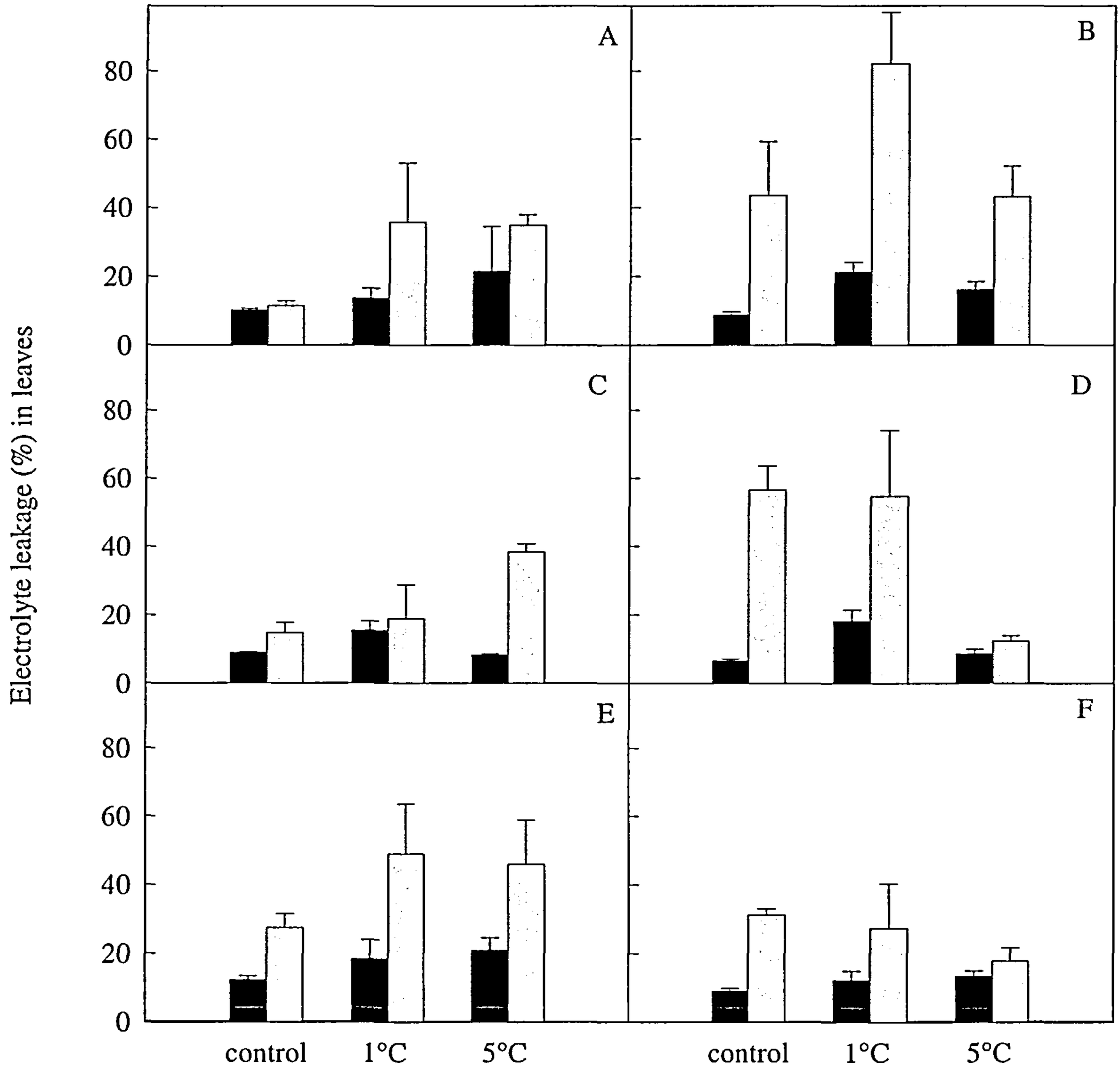


Figure 5.8: Electrolyte leakage (%) in leaves of 'First Red' (A, C, E) and 'Akito' (B, D, F) roses on days 0 (■) and 10 (□) of vase life. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were put into vases without ABA (control) (A, B) or sprayed (C, D) or pulsed (E, F) with ABA. Data are independent treatment means (± S.E.); n = 12. Main factor means are presented in Table 5.4. ANOVA and Duncan's tests are presented in Appendix 4.5.1, Tables A5.5.1.6 – A5.5.1.10.

Lipid peroxidation in petals and leaves

After 10 days of vase life, there was a significant increase in malondialdehyde (MDA) content in petals and leaves, suggesting that lipid peroxidation was increasing during vase life evaluation (Table 5.5, Figures 5.9 and 5.10). The levels of MDA were markedly higher in 'First Red' than in 'Akito' roses by *ca.* 7.39- and 2.27-fold in petals and leaves, respectively as a mean of all treatments.

Storing roses at 1 and 5°C significantly increased MDA levels in petals and leaves of both cultivars (Table 5.5, Appendix 5.5.2, Tables A5.5.2.1 – A5.5.2.12). MDA content in 'First Red' roses at 1°C did not differ from that at 5°C. Main factor means indicated that when 'Akito' roses were stored at 1°C, MDA content in leaves significantly increased by *ca.* 1.78- and 1.55-fold on days 0 and 10, respectively (Table 5.5, Appendix 5.5.2, Tables A5.5.2.9 and A5.5.2.11). Increased MDA content in 'Akito' leaves at 1°C was more pronounced after pulsing with ABA (Figure 5.10B, D, F, Appendix 5.5.2, Tables A5.5.2.10 and A5.5.2.12). ABA treatments did not affect the degree of lipid peroxidation in leaves and petals of 'First Red' and in petals of 'Akito' roses, respectively. Main factor means indicated a significant increase in MDA in leaves of 'Akito' roses caused by pulse ABA treatment on day 0 (Table 5.5). However, after 10 days of vase life, there was no significant effect of ABA treatments on MDA content in leaves for 'Akito' roses.

Table 5.5: Effects of storage and ABA treatments on malondialdehyde (MDA; mM^{-1} cm^{-1}) content in petals and leaves of 'First Red' and 'Akito' roses on days 0 and 10. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were pulsed or sprayed with ABA or put into vases without ABA (control). Data are main-factor $\bar{x}$; n = 36. Data for independent treatment means are presented in Figures 5.9 and 5.10. ANOVA and Duncan's tests are presented in Appendix 5.5.2.

Factors	'First Red'		'Akito'	
	Day 0 [a]	Day 10	Day 0	Day 10
	a. MDA (mM^{-1} cm^{-1}) in petals			
1) Storage treatment [b]				
control	87.5a	196.8a	22.1a	23.9a
1°C	175.2b	213.0a, b	28.1b	26.3b
5°C	219.1b	257.8b	26.9b	28.3b
2) ABA treatment				
control	144.1a	199.1a	25.4a	27.4a
spray	148.9a	220.9a	26.5a	27.6a
pulse	188.5a	247.5a	25.2a	26.5a
	b. MDA (mM^{-1} cm^{-1}) in leaves			
1) Storage treatment				
control	59.3a	71.7a	33.0a	38.7a
1°C	126.4b	140.6b	59.0b	60.3b
5°C	117.8b	136.2b	45.3c	50.4c
2) ABA treatment				
control	97.8a	109.7a	42.4a	47.3a
spray	103.3a	120.3a	43.3a, b	47.4a
pulse	102.3a	118.4a	51.5b	54.7a

[a] Data are main factor means of MDA in petals and leaves. [b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

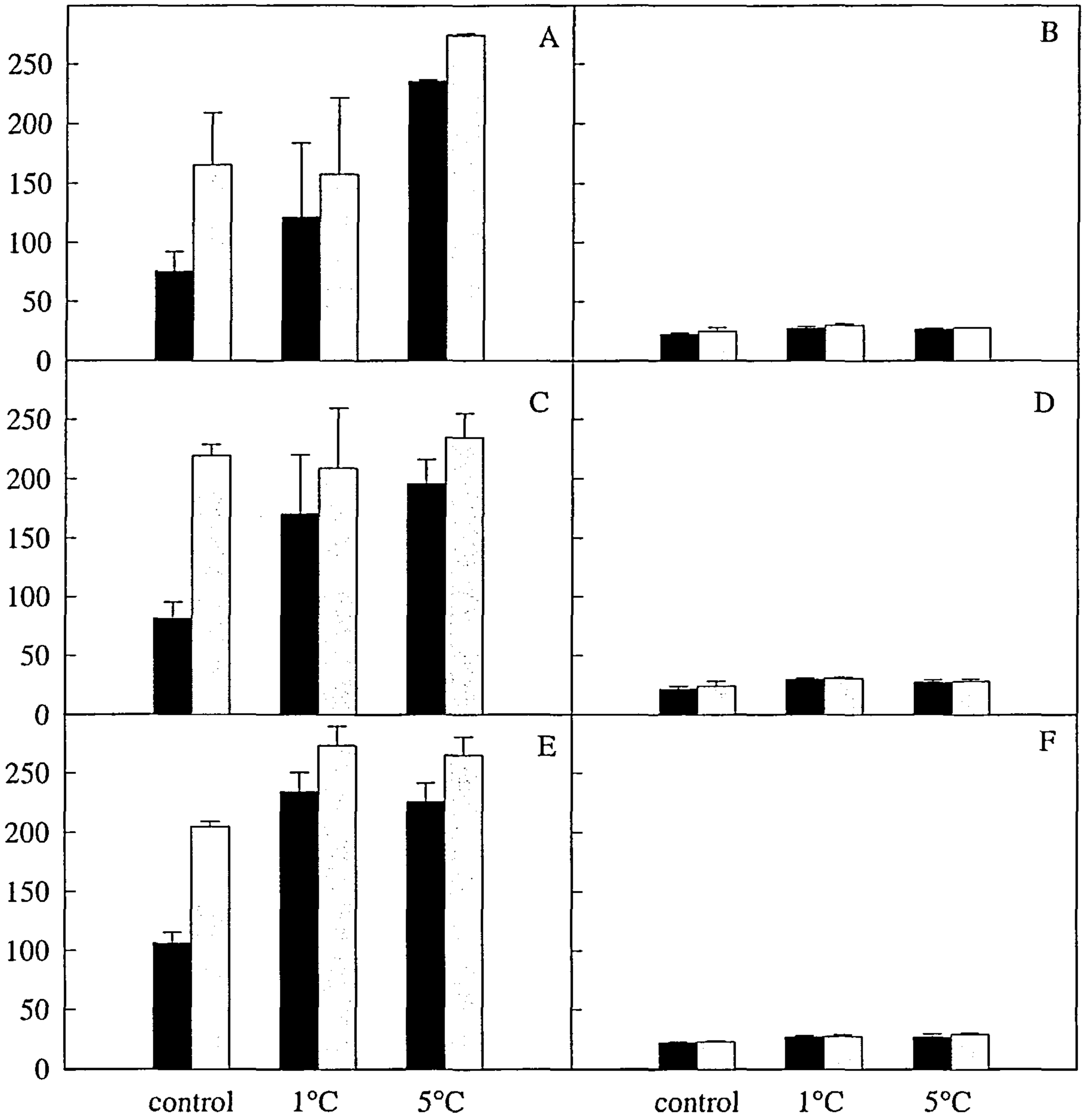


Figure 5.9: Malondialdehyde (MDA; mM^{-1} cm^{-1}) content in petals of 'First Red' (A, C, E) and 'Akito' (B, D, F) roses on days 0 (■) and 10 (□) of vase life. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were put into vases without ABA (control) (A, B) or sprayed (C, D) or pulsed (E, F) with ABA. Data are independent treatment means (± S.E.); n = 12. Main factor means are presented in Table 5.5. ANOVA and Duncan's tests are presented in Appendix 5.5.2, Tables A5.5.2.1 – A5.5.2.5.

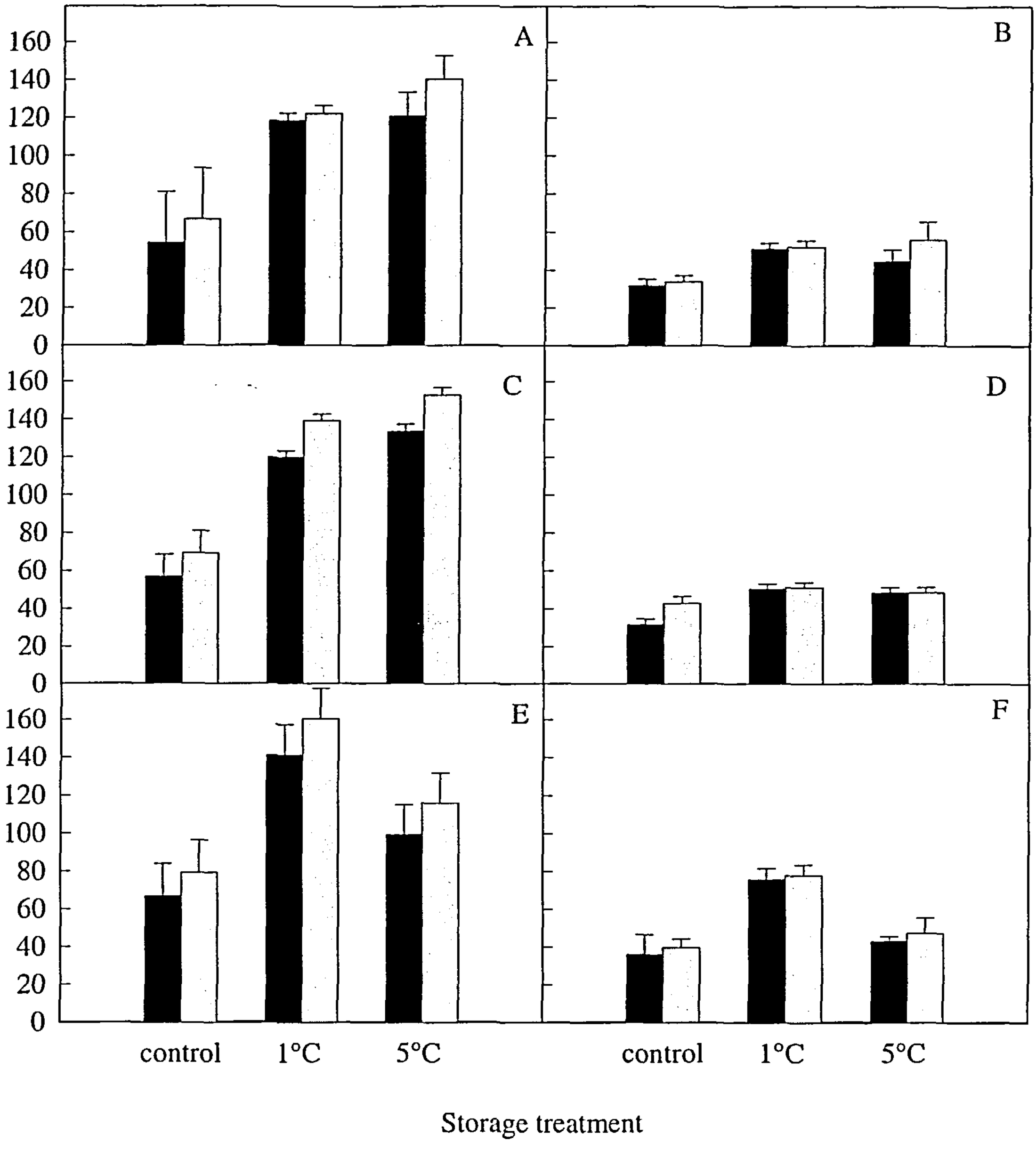


Figure 5.10: Malondialdehyde (MDA; mM^{-1} cm^{-1}) content in leaves of 'First Red' (A, C, E) and 'Akito' (B, D, F) roses on days 0 (■) and 10 (□) of vase life. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were put into vases without ABA (control) (A, B) or sprayed (C, D) or pulsed (E, F) with ABA. Data are independent treatment means (± S.E.); n = 12. Main factor means are presented in Table 5.5. ANOVA and Duncan's tests are presented in Appendix 5.5.2, Tables A5.5.2.6 – A5.5.2.12.

Endogenous ABA content in petals and leaves

ABA content (ng g^{-1} f.w.) in leaves of 'Akito' roses was *ca.* 8.8-fold greater than that in petals as a mean for all treatments (Table 5.6, Figures 5.11 and 5.12). ABA content in leaves markedly increased from day 0 to 10 (Figure 5.12). The same increase in ABA content was recorded in petals during vase life, except from those sprayed with ABA and then stored at 1 and 5°C (Figure 5.11). In these flowers, ABA content in petals did not change from day 0 to 10. Storage of roses at 5°C significantly ($P < 0.05$) increased endogenous ABA production by *ca.* 7.2-fold in leaves and *ca.* 1.15-fold in petals on days 10 and 0, respectively (Table 5.6, Appendix 5.5.3, A5.5.3.2 and A5.5.3.4). Main Factor means suggested that pulsing roses with 10^{-1} M ABA for 24 hours resulted in increased ABA content in leaves and petals (Table 5.6, Appendix 5.5.3, A5.5.3.2 and A5.5.3.4). Duncan's multiple range tests indicated that when roses were pulsed with ABA and then stored at 5°C, ABA levels in leaves and petals on days 0 and 10, respectively, were greatest compared to other treatments (Figures 5.11 and 5.12, Appendix 5.5.3, Tables A5.5.3.3 and A5.5.3.5).

Table 5.6: Effects of storage and ABA treatments on ABA content (ng g^{-1} f.w.) in petals and leaves of 'Akito' roses on days 0 and 10. Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were pulsed or sprayed with ABA or put into vases without ABA (control). Data are main-factor $\bar{x}$; n = 9. Data for independent treatment means are presented in Figures 5.11 and 5.12. ANOVA and Duncan's tests are presented in Appendix 5.5.3.

Factors	**ABA content in petals**		**ABA content in leaves**	
	Day 0 [a]	**Day 10**	**Day 0**	**Day 10**
1) Storage treatment [b]				
control	29.1a	129.8a	84.7a	1283.2a
1°C	29.8a	150.8a	102.3a	1267.5a
5°C	39.4a	267.0b	616.4b	2369.7a
2) ABA treatment				
control	20.6a	144.5a	189.1a	1307.1a
spray	32.7a, b	82.8a	224.7a, b	1563.2a
pulse	44.9b	320.3b	389.6b	2050.0a

[a] Data are main factor means of ABA in petals and leaves. [b] Within main factor means, numbers followed by the same letter are not significantly different at P = 0.05.

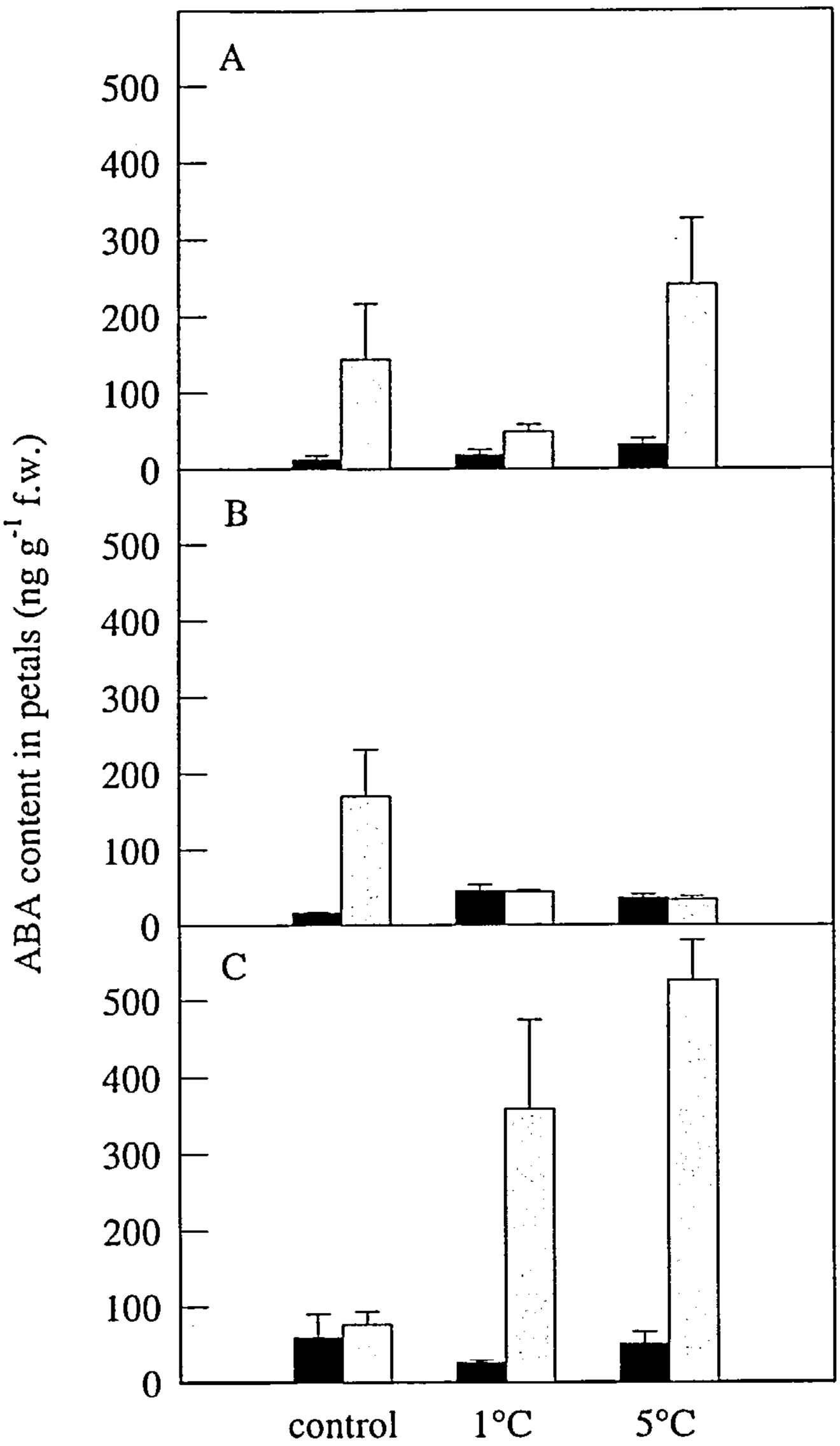


Figure 5.11: ABA content (ng g^{-1} f.w.) in petals of 'Akito' roses on days 0 (■) and 10 (▩). Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were put into vases without ABA (control) (A) or sprayed (B) or pulsed (C) with ABA. Data are independent treatment means (± S.E.); n = 3. Main factor means are presented in Table 5.6. ANOVA and Duncan's tests are presented in Appendix 5.5.3, Tables A5.5.3.1 – A5.5.3.3.

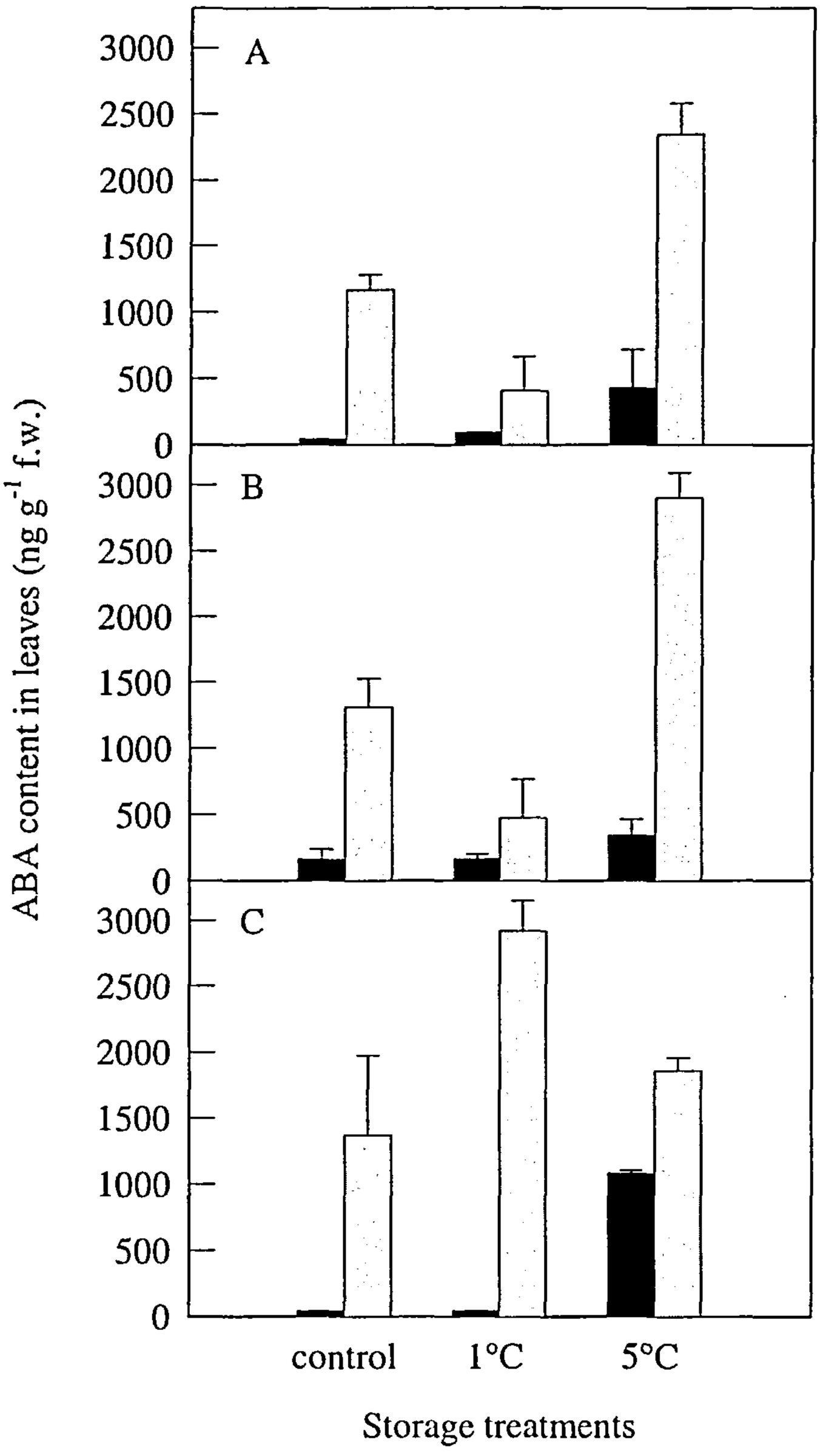


Figure 5.12: ABA (ng g^{-1} f.w.) content in leaves of 'Akito' roses on days 0 (■) and 10 (□). Flowers were not stored (control) or stored wet at 1 and 5°C for 10 days. Before storage, flowers were put into vases without ABA (control) (A) or sprayed (B) or pulsed (C) with ABA. Data are independent treatment means (± S.E.); n = 3. Main factor means are presented in Table 5.6. ANOVA and Duncan's tests are presented in Appendix 4.5.3, Tables A5.5.3.4 – A5.5.3.6.

5.2.1.6 Relationship between vase life duration and vase life parameters

A weak but significant association ($P < 0.05$, $r = 0.44$) was found between leaf F_v/F_m value on day 0 of vase life and flower longevity of 'First Red' roses under the conditions of these experiments (Table 5.7). Moreover, flower life of 'First Red' was positively correlated with fresh weight ($P < 0.05$, $r = 0.45$) and solution usage ($P < 0.01$, $r = 0.49$) on days 8 and 9, respectively. Similarly, foliage life was significantly correlated with fresh weight on day 8 ($P < 0.01$, $r = 0.67$) and solution usage on day 9 ($P < 0.01$, $r = 0.55$). Increasing the rate of electrolyte leakage in leaves of 'First Red' roses was significantly correlated ($P < 0.05$, $r = -0.45$) with shortening of foliage life. Negative correlations between MDA content in leaves and foliage life were also determined for 'First Red' ($P < 0.05$, $r = -0.46$) and 'Akito' roses ($P < 0.05$, $r = -0.49$). These associations may indicate the detrimental effects of lipid peroxidation in leaves on the subsequent vase life.

5.2.1.7 Glasshouse environmental conditions

In the present study, vase life experiments were carried out using roses grown from September 2003 to February 2004 (Chapter 3, section 3.3.1). Mean day and night temperatures were greater in September and October followed by those in February and November (Figure 5.13A). Mean day and night minimum temperatures were recorded in December and January. Similar changes in growing temperature were recorded in autumn and winter months the first year (Chapter 4, section 4.2.2.1). However, both day and night temperatures were greater in second than in first year. Mean day and night RH increased from September to January (Figure 5.13B) due to the progressive decline of temperature. In January, RH reached maximum values of *ca.* 95 and 75% in night and day, respectively. PFD was progressively decreased from September to January (Figure 5.13C). In autumn and winter months, PFD of second year was comparatively greater than that of first year (Chapter 4, section 4.2.2.1). For example, PFD reached *ca.* 1750 μmol m^{-2} s^{-1} in September of second year, while it did not pass 1500 μmol m^{-2} sec^{-1} in September of first year.

Table 5.7: Effect of flower and foliage lives on vase life parameters (e.g. F_v/F_m on d 0, electrolyte leakage, MDA, corolla diameter on d 8, fresh weight on d 8 and solution usage on d 7) of 'First Red' and 'Akito' roses grown from autumn 2003 to winter 2003 – 04. Flowers were not stored or stored wet at 1 and 5°C. Before storage, flowers were pulsed or sprayed with ABA or put into vases without ABA (control).

Vase life variables	Flower life [a]		Foliage life	
	'First Red'	'Akito'	'First Red'	'Akito'
F_v/F_m (d 0)	0.44 *	0.01 n.s.	0.29 n.s.	0.05 n.s.
Electrolyte leakage [b]	0.05 n.s.	-0.05 n.s.	-0.45 *	-0.23 n.s.
MDA [b]	-0.31 n.s.	-0.07 n.s.	-0.46 *	-0.49 *
Corolla diameter (d 8)	0.32 n.s	-0.09 n.s.	0.15 n.s.	0.19 n.s.
Fresh weight (d 8)	0.45 *	0.33 n.s.	0.67 **	0.21 n.s.
Solution usage (d 7)	0.49 **	-0.02 n.s.	0.55 **	-0.15 n.s.
ABA content [b]		0.06 n.s.		0.06 n.s.

[a] Data are results from Pearson's correlation at P = 0.05; n = 108. [b] Electrolyte leakage, MDA and ABA content are means of days 0 and 10 of vase life. ** Significance at P = 0.01, * Significance at P = 0.05, n.s. not significant at P = 0.05.

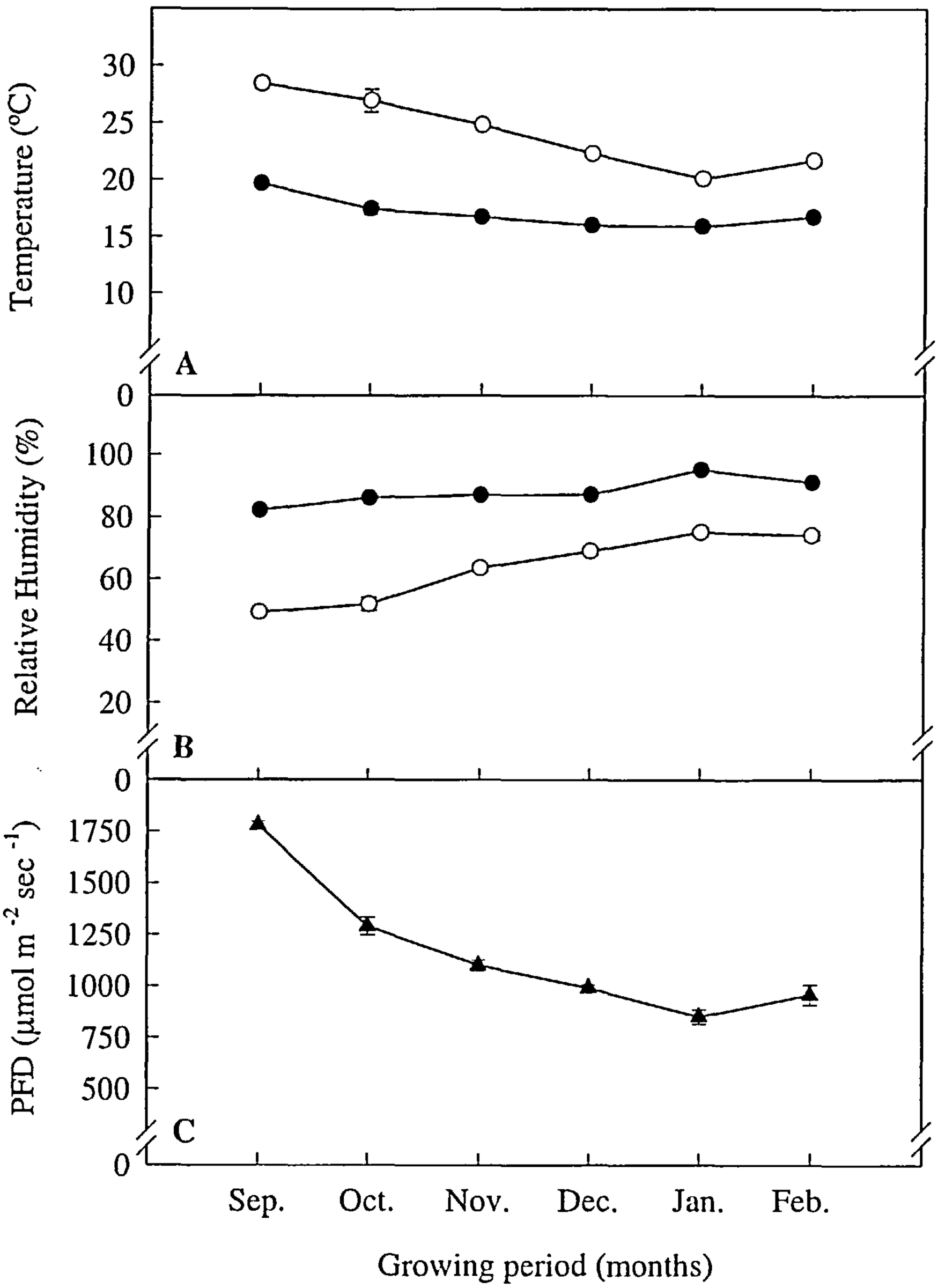


Figure 5.13: Changes in (A) temperatures (°C) and (B) RH (%) during the day (○) and night (●), and (C) Photon Flux Density (PFD) (µmol m^{-2} s^{-1}) in the glasshouse environments throughout the growing period (from September 2003 to February 2004). Data are means of 12 recordings per day; *n* = number of days each month. Vertical bars show ± SE for each month (*n* = number of days each month).

5.2.2 Effects of growing season on cell lignification of 'First Red' and 'Akito' roses.

Increased sensitivity of 'Akito' roses to bent neck compared to 'First Red', previously reported here (Chapter 5, section 5.2.1.3 and Chapter 4, section 4.2.1.2), was apparently due to the low number of vascular bundles with lignin on cell walls (Table 5.8, Appendix 5.5.4, Tables A5.5.4.1 and A5.5.4.2). The number of xylem elements with lignified cells per vascular bundle was significantly lower in 'Akito' than in 'First Red' roses (Appendix 5.5.4, Table A5.5.4.4). Increased number of lignified cells in 'Akito' compared to 'First Red' roses was clearly visible either under cool light illuminator after staining with phloroglucinol-HCl (Plate 5.1) or under fluorescence light illuminator after staining Safranin O and counterstaining in Fast Green (Plate 5.2). Growing both cultivars in winter significantly ($P \leq 0.001$) decreased the number of vascular bundles in the peduncle and the lignified xylem elements per vascular bundle compared to autumn-grown roses (Appendix 5.5.4, Tables A5.5.4.3 and A5.5.4.4).

Growing season did not affect vascular bundle diameter in peduncles, suggesting similarity in size of vascular bundles for roses grown in different seasons (Table 5.9, Appendix 5.5.4, Tables A5.5.4.5 and A5.5.4.7). In winter experiments, vascular bundle diameter in peduncles was 0.353 and 0.317 mm for 'First Red' and 'Akito' roses, respectively. Analysis of variance showed that this difference was significant ($P < 0.05$). In autumn, however, there were no significant differences between cultivars. The total area (%) of vascular bundle in transverse section was significantly less in 'First Red' compared to 'Akito' roses (Table 5.9, Appendix 5.5.4, Tables A5.5.4.6). Thus, although 'First Red' roses had greater number of vascular bundles (Table 5.8), their total area (%) in peduncles did not exceed the area of vascular bundles in 'Akito' peduncles. This finding is due to the lower diameter of 'Akito' peduncles, which resulted in increased total area of vascular bundles. Vascular bundle area in peduncles of both cultivars was not affected by growing season.

Table 5.8: Number of vascular bundles and xylem elements per vascular bundle measured in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter after staining with phloroglucinol-HCl for lignified cell walls. Data are independent treatment means; n = 9. ANOVA and Duncan's tests are presented in Appendix 5.5.4, Tables A5.5.4.1 – A5.5.4.4.

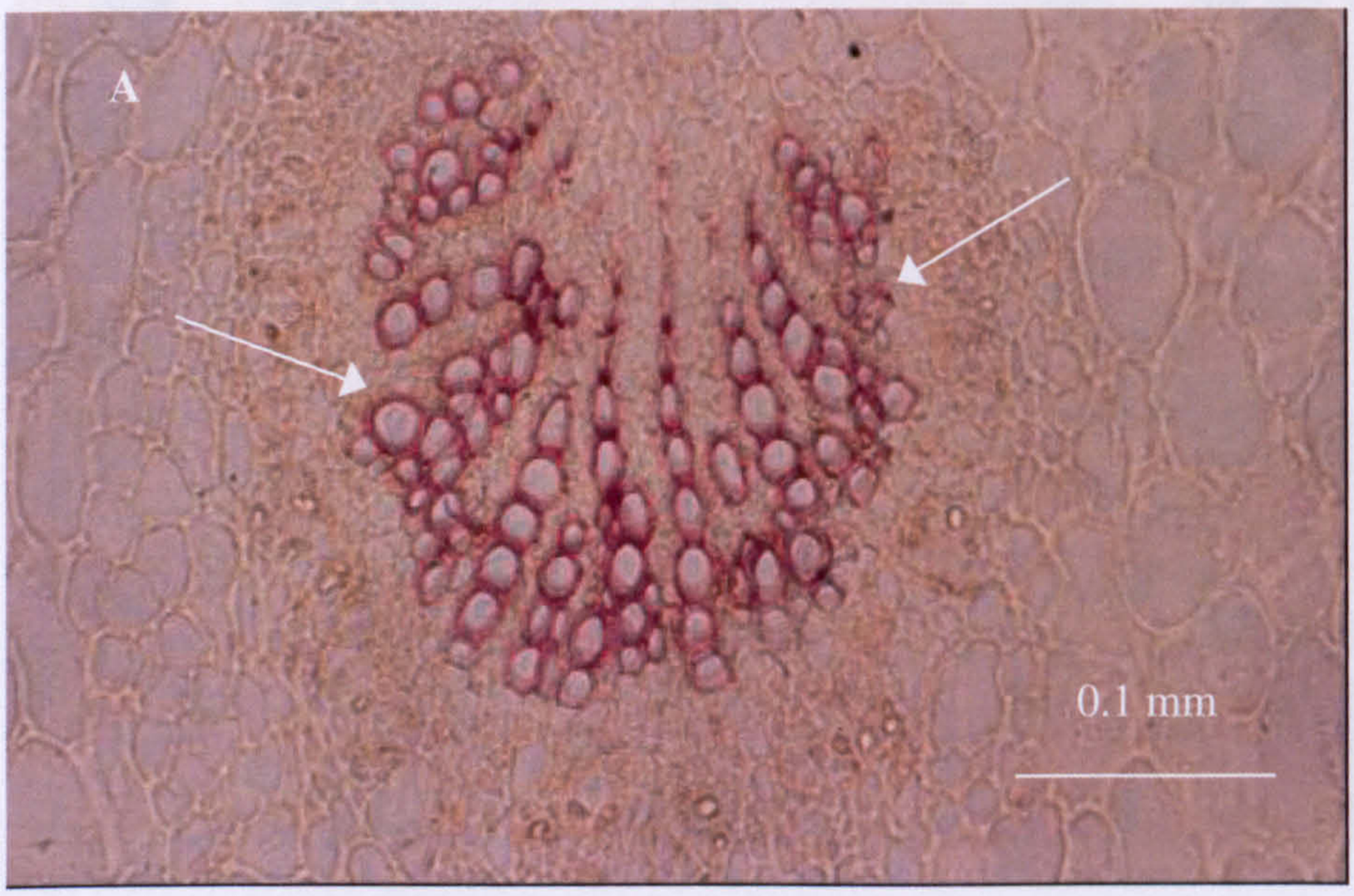
Treatments	Number of vascular bundles	Xylem elements / Vascular bundle [b]
a. Autumn experiments		
'First Red' [a]	66.8a	59.7a
'Akito'	43.0b	53.3b
b. Winter experiments		
'First Red'	59.8c	68.8c
'Akito'	36.0d	41.0d

[a] Numbers followed by the same letter within column are not significantly different at P = 0.05. [b] Xylem elements data are means of 10 random vascular bundles in each transverse section.

Table 5.9: Mean diameter of vascular bundle (mm) and vascular bundle area (%) of transverse section measured in 'First Red' and 'Akito' peduncles in autumn and winter after staining with phloroglucinol-HCl for lignified cell walls. Data are independent treatment means; n = 9. ANOVA and Duncan's tests are presented in Appendix 5.5.4, Tables A5.5.4.5 – A5.5.4.8.

Treatments	Vascular bundle diameter (mm)	Vascular bundle area (% of transverse section)
a. Autumn experiments		
'First Red' [a]	0.368a	4.5a
'Akito'	0.336ab	5.5b
b. Winter experiments		
'First Red'	0.353a	4.3a
'Akito'	0.317b	5.2b

[a] Numbers followed by the same letter within column are not significantly different at P = 0.05.



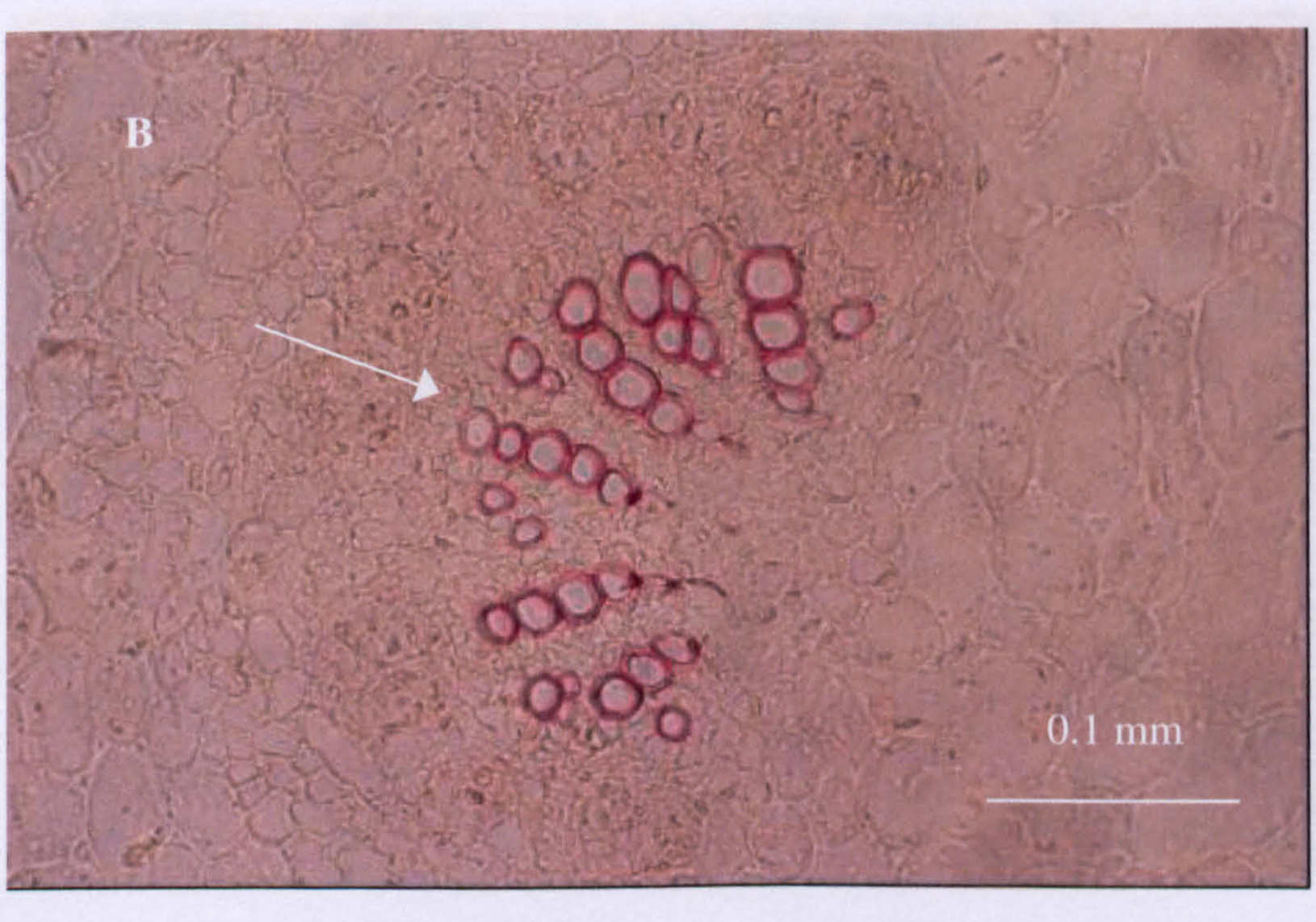
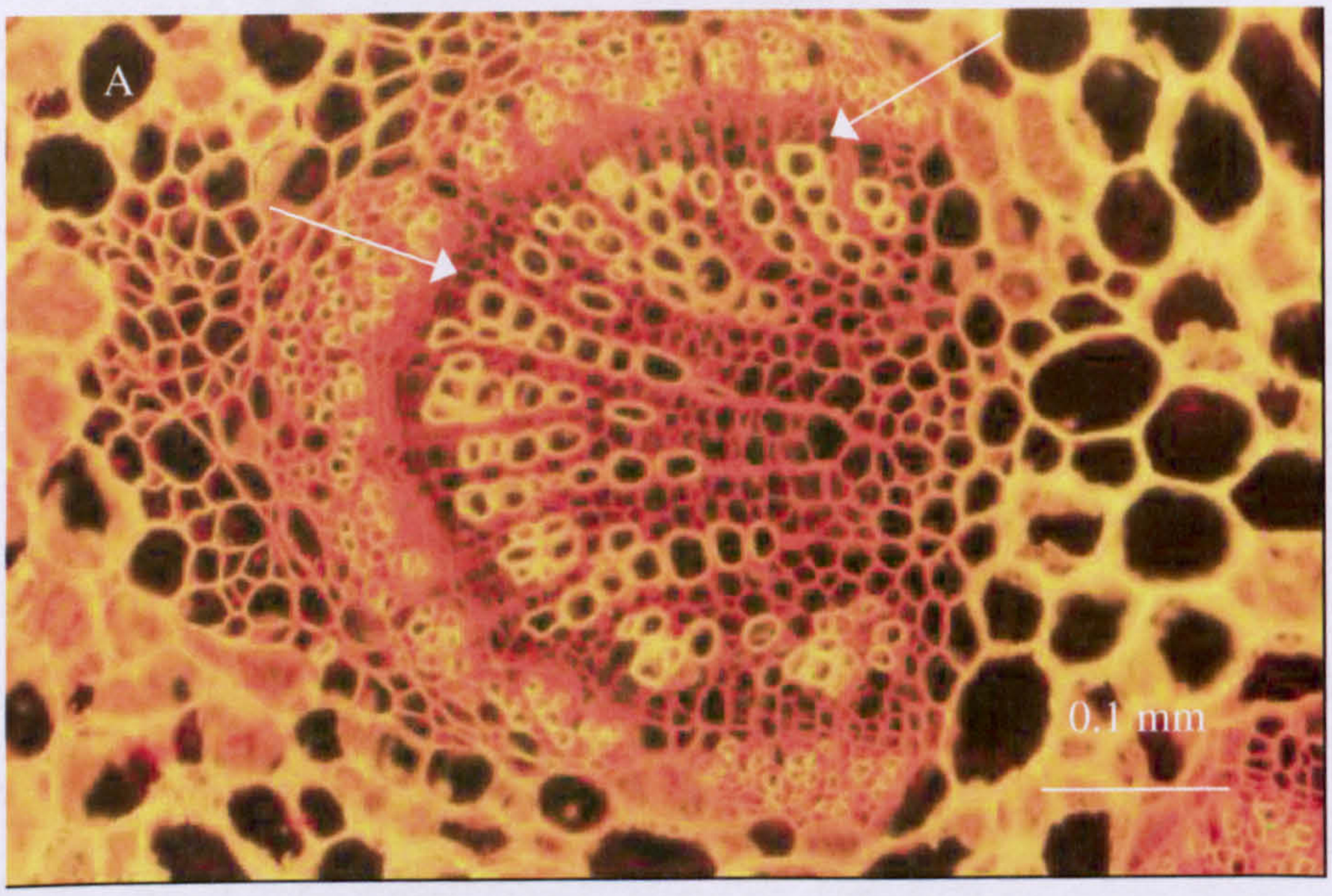


Plate 5.1: Transverse section of 'First Red' (A) and 'Akito' (B) vascular bundle consisting of xylem cells. Within vascular bundle, lignified cell walls (arrow) were stained with phloroglucinol-HCl (Zhong *et al.*, 2000) and examined under cool light illuminator.



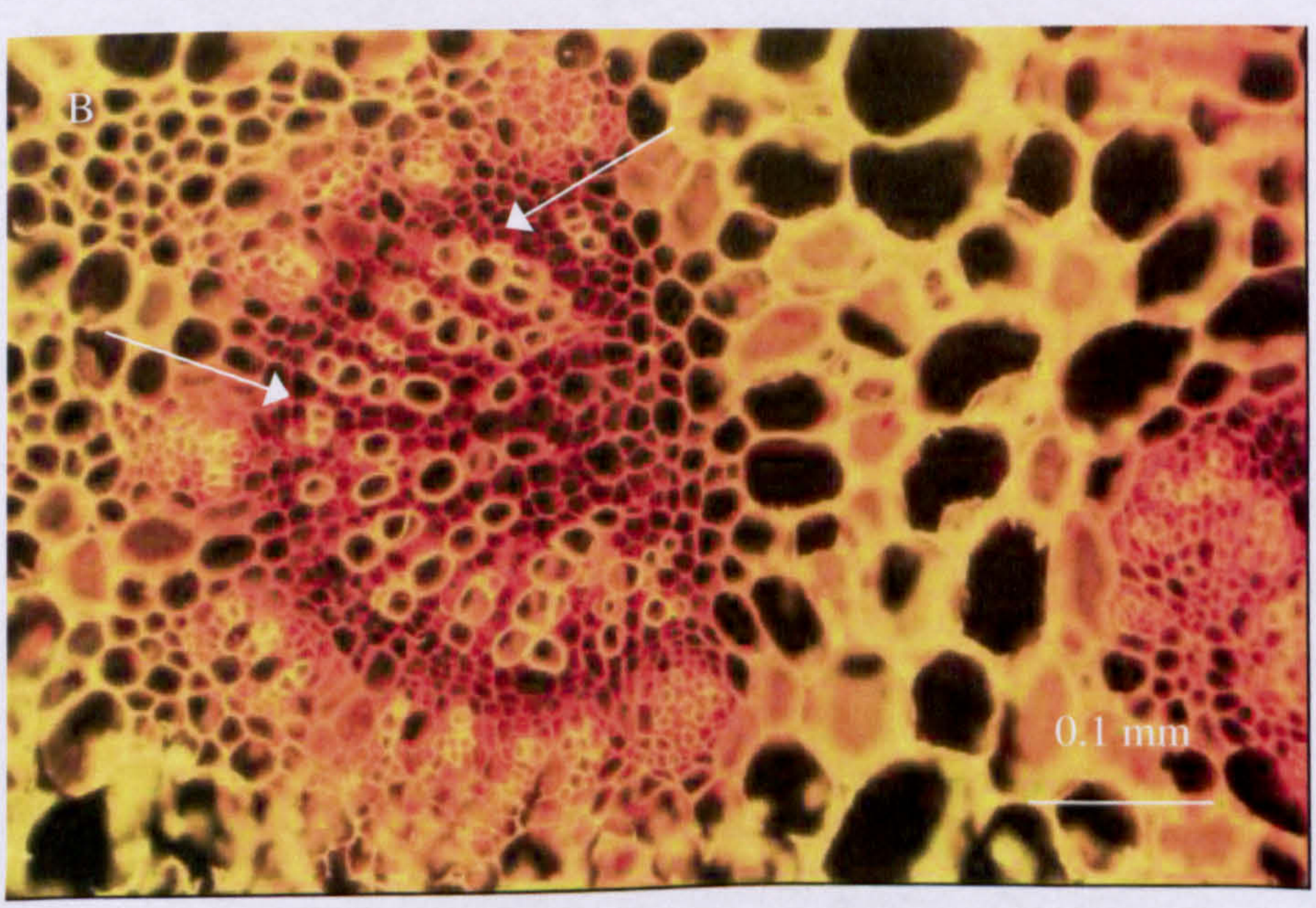


Plate 5.2: Transverse section of 'First Red' (A) and 'Akito' (B) vascular bundle consisting of xylem cells. Within vascular bundle, fluorescing tissue under UV-light at 340-380 nm (arrows) indicates deposition of lignin and callose on cell walls of xylem elements (Ruzin, 1999).

5.3 DISCUSSION

5.3.1 Effects of storage temperature on vase life parameters of roses grown in autumn and winter

Electrolyte leakage and MDA (e.g. lipid peroxidation) have long been used as indicators of damage to the plasma membrane and LTI or CI in plant tissue (Heath and Packer, 1968; Meir *et al.*, 1992; Zhuang *et al.*, 1995). MDA production in 'First Red' was significantly greater than in 'Akito' roses for all treatments, indicating that the degree of lipid peroxidation in roses is strongly affected by genotype. Both electrolyte leakage and MDA were increased during vase life suggesting accelerated senescence (Marangoni *et al.*, 1996) and/or senescence-associated changes in peroxide-scavenging activity (Sung and Jeng, 1994).

Storing 'Akito' roses at 1°C for 10 days significantly reduced foliage life enhancing MDA production (e.g. lipid peroxidation) and electrolyte leakage in leaves. Similarly, Murata (1989) reported similar drastic increases in ion leakage in cucumber, melon and winter squash stored at 0°C for 8, 15 and 28 days, respectively. Increased lipid peroxidation and electrolyte leakage have also been found in tomato fruit during and after chilling (Sharom *et al.*, 1994). These findings have been attributed to chilling-induced alterations in cell membrane properties that can alter functionality (Marangoni *et al.*, 1996). During lipid degradation by peroxidation, unsaturated fatty acids yield hydroperoxides, which are further degraded to water-extractable aldehydes, such as malondialdehydes (MDA) (Meir *et al.*, 1992). MDA is an active compound, which has been found to further react with proteins, phospholipids and nucleic acids, thereby creating lipofuscin-like compounds (Malshet and Tappel, 1973). These changes induce modifications of membrane permeability and alterations of enzyme activities (Chapter 3, section 2.2.1.1). In the current experiments, increasing MDA content in leaves of 'First Red' and 'Akito' roses was significantly correlated with foliage life decline. Therefore, increased lipid peroxidation in leaves of roses was associated with reduced foliage life.

Flower and foliage lives of 'First Red' roses were significantly reduced after storage at 1 or 5°C compared to non-stored control flowers. Similarly, 'First Red' roses stored at 1 or 5°C had less capacity in maintaining fresh mass and taking up vase solution during vase life. Both fresh weight and solution usage by 'First Red' roses

on day 8 and 7, respectively, were positively correlated with vase life. These significant correlations suggest that fresh weight measured on day 8 and vase solution usage by flowers from day 6 to 8 are potentially useful indices of flower and foliage vase lives (Pompodakis *et al.*, 2004). Moreover, there was evidence of vase life decline in 'First Red' roses with increasing storage temperature from 1 to 5°C. In Chapter 4, the reductions in vase life of roses with increasing storage temperature from 1 to 10°C were attributed to the developed senescence during storage (section 4.3.2). In the current experiments, this explanation is strongly supported for 'First Red' roses by the results of electrolyte leakage and MDA in leaves and petals, respectively. Both parameters were increased with increasing storage temperature from 1 to 10°C and were negatively correlated with vase life duration, indicating that increased membrane permeability and lipid peroxidation are related to vase life reductions (Faragher *at al.*, 1986; Rubinstein, 2000).

Reducing storage temperature of 'First Red roses was associated with reduction in F_v/F_m value, indicating LTI effects on PSII activity, as discussed in Chapter 4. Similar reductions in F_v/F_m value were reported for 'Akito' roses grown in winter and then stored at 1°C in Chapter 4, suggesting increased sensitivity of this cultivar to LTI (section 4.3.2). However, under the conditions of current experiments, storage temperature did not alter F_v/F_m of 'Akito' roses. As it has been discussed earlier in Chapter 4, the reduction of F_v/F_m at 1°C in roses was not only due to direct effects of storage temperature but was due to an interactive effect of storage temperature and growing season. Indeed, F_v/F_m changes after storage of 'Akito' roses were strongly associated with changes in PFD during the year (Chapter 4, section 4.2.2.2). Thus, seasonal variation could strongly affect F_v/F_m value after low temperature storage. In the present study (second year), growing temperature and PFD in autumn and winter months were comparatively greater than those of chapter 4. Thus, the greater F_v/F_m in 'Akito' roses may be attributed to the greater PFD and/or growing temperature. However, in both first and second year, F_v/F_m in 'First Red' roses declined with reducing storage temperature (Chapter 4, section 4.2.1.1 and Chapter 5, section 5.2.1.2). This reproducible finding for 'First Red' roses indicates that the fall of F_v/F_m is stronger affected by storage temperature rather than seasonal variation.

In most cases, storage of roses increased endogenous ABA levels. When 'Akito' roses were stored at 5°C, significant ($P < 0.05$) increases of *ca.* 7.2-fold in

leaves and *ca.* 1.15-fold in petals were detected on days 10 and 0, respectively. An elevated level of endogenous ABA caused by water stress has been reported previously for roses (Orlandini, 1991) and petunias (Vardi and Mayak, 1989). The developed water stress stimulated senescence in petunia flowers through ABA production. Thus, a possible development of water stress, which is common for roses during storage (Hu *et al.*, 1998a, b), could affect ABA levels during vase life (Dallaire *et al.*, 1994). Our results, which have not been previously reported for roses after storage, are also in agreement with work on chilling sensitive species reported above (section 5.1). At low temperature, the capacity of these species to produce ABA is linked to ABA protective mechanisms (section 5.1).

5.3.2 Effects of ABA treatments on vase life parameters

ABA applied in 'Akito' roses before storage at 5°C extended both flower and foliage lives. Pulsing or spraying 'First Red' roses with ABA also increased foliage life of controls and flowers stored at 5°C. Such extensions of rose longevity by exogenous ABA application have been attributed to ABA accumulation in guard cells and induction of stomatal closure (Wilkinson and Davies, 2002; Pompodakis and Joyce, 2003). Indeed, before storage at 5°C, pulse treatment increased ABA content in leaves and petals on days 0 and 10, respectively. Pulsing 'Akito' roses with ABA reduced the rate of solution usage the first days of vase life, supporting that applied ABA evidently caused stomatal closure. Additionally, water stress during storage could affect active ABA concentrations in the apoplast of guard cells, with sap alkalisation enhancing the physiological activity of ABA (Hartung *et al.*, 1998; Davies *et al.*, 2002). Spraying 'First Red' roses with ABA prevented F_v/F_m decline on day 0 of vase life. This positive effect of ABA on F_v/F_m maintenance may be related to the protective role of ABA against LTI and/or water deficit stress (section 5.3.1).

Significant increases in ABA content were detected in petals and leaves of 'Akito' roses after 10 days of vase life compared to day 0. ABA content in rose petals has been found to decrease during the first 3 days of vase life (Le Page-Degivry *et al.*, 1991), followed by a steady state at low levels, and finally by a sharp increase in the last stages of senescence (Borochov *et al.*, 1976b). The first step (e.g. ABA decline) was not recorded in our experiments, as ABA was not measured during the first days of vase life. However, the second step (e.g. sharp ABA increase) was recorded in

'Akito' petals and leaves on day 10 of vase life. This second step (e.g. ABA increase) might be in parallel with ethylene production in petals (Hanley and Bramlage, 1989; Muller *et al.*, 1999; Sood and Nagar, 2003) and with changes in water status of leaves. This tendency of ABA to increase during advanced stages of senescence was also observed in senescing petals of carnation (Hanley and Bramlage, 1989), in detached senescing leaves of tobacco (Even-Chen and Itai, 1975) and rice leaves (Philosoph-Hadas *et al.*, 1993).

5.3.3 Histological study of bent neck

First year results indicated great sensitivity of 'Akito' roses to bent neck (Chapter 4, section 4.2.1.2). The symptom was recorded the last days of vase life during the advanced stages of senescence and, thereby, did not affect vase life duration. In the present study, however, the increased bent neck of 'Akito' roses was recorded the first days of vase life resulting in flower life reductions (personal observations). Bent neck in roses occurs under water stress conditions and/or when maturation and lignification of the peduncle is not complete (Zieslin *et al.*, 1978). Increased transpiration rates (Marousky, 1969; Carpenter and Rusmussen, 1975) and the development of vascular occlusions within the flower stem (Woltering, 1987; Bleeksma and van Doorn, 2003) can cause water stress resulting in bent neck problems. Moreover, the rigidity of the peduncle is of particular importance (Burdett, 1970). Histological studies in transverse sections of 'Akito' peduncles showed that lignified vascular bundles and xylem elements per vascular bundle were significantly less in the peduncle of 'Akito' compared to 'First Red' roses. Similar differences in xylem elements between rose cultivars have been found to affect the capacity of the xylem cell walls for transport of water (van Doorn and Reid, 1995). Both 'First Red' and 'Akito' peduncles had less lignified cells, when they were grown in winter. The low degree of lignification during winter is apparently related to the lower supply of photosynthetic products, lower levels of IAA, and decreased peroxidase activity (Parups and Voisey, 1976).

CHAPTER 6

EFFECT OF ABA AND ABA ANALOGUE TREATMENTS, BEFORE AND AFTER STORAGE AT 1°C, ON VASE LIFE OF CUT 'AKITO' ROSES

6.1 INTRODUCTION

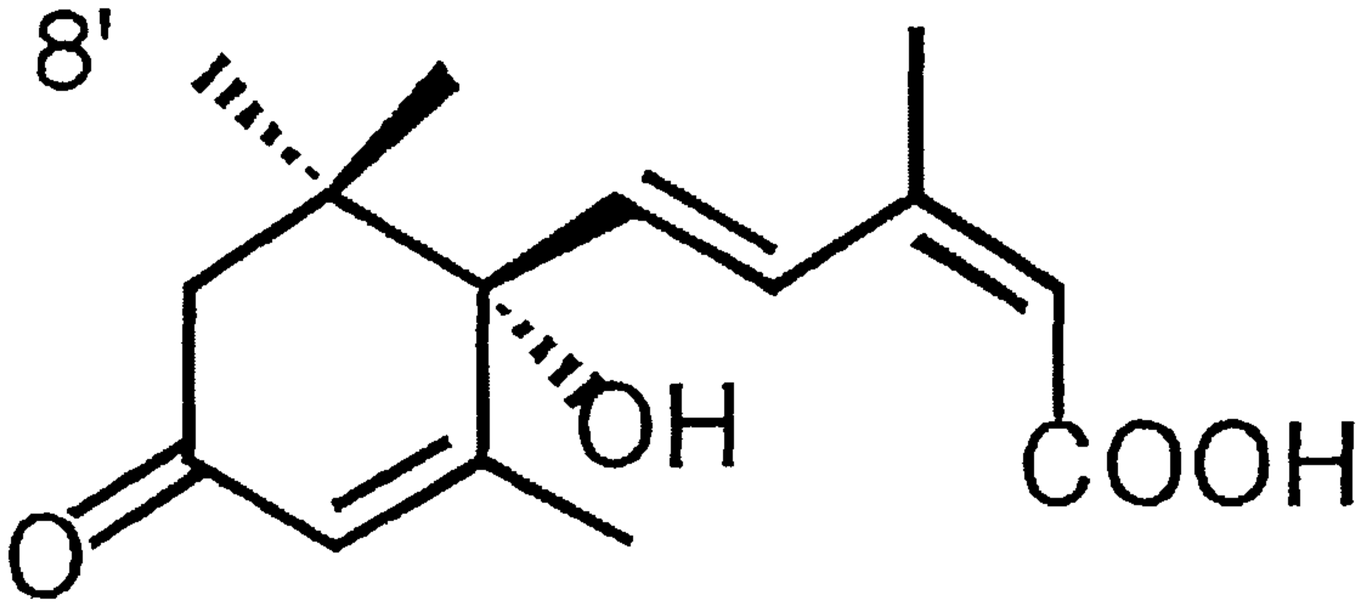
Abscisic acid (ABA) can cause physiological responses that protect cut flowers against CI or LTI (Chapter 4, section 5.1). However, ABA from 'fine-chemical' companies is expensive and ABA solutions can be unstable due to photodegradation (Joyce *et al.*, 1996). Comparatively inexpensive alternatives are required for use by the cut flower industry. The cost of ABA analogues is about 15-fold less expensive than that of pure ABA. Thus, synthetic ABA analogues could be useful.

Recently, novel ABA analogues have been synthesised by Canada Agriculture (Plant Biotechnology Institute, Saskatoon, Canada). These compounds are sterically almost identical to pure ABA (Figure 6.1). ABA analogues (e.g. 8′-methylene ABA) are perceived as ABA-like by plants but are turned over much more slowly resulting in enhanced biological activity (Lamb *et al.*, 1996; Rose *et al.*, 1997). ABA analogues interact in various physiological plant processes, including stress avoidance mechanisms that reduce water loss (Abrams *et al.*, 1997). Addition of synthetic ABA analogues PBI-365 and PBI-429 into vase solution reduced solution usage by 'Bacarra' roses (Pompodakis and Joyce, 2003), possibly by inducing stomatal closure (Trejo *et al.*, 1993). Moreover, like ABA, the analogue PBI-365 reduced leaf crisping (Pompodakis and Joyce, 2003; Chapter 2, section 2.2.4), which had been caused by increased sucrose concentration in the leaf apoplast (Markhart and Harper, 1995; Torre and Fjeld, 2001).

The aim of these experiments was to investigate the effect of ABA and synthetic ABA analogue (e.g. PBI-365) on chilling tolerance of roses. Cut roses were pulsed or not (control) with 10^{-1} M ABA for 24 hours and then stored at 1°C for 10 days. After storage, roses were placed in vase solutions containing 10^{-5} M ABA, 10^{-5} M PBI-365 or distilled water (control). Vase life parameters, (e.g. vase life duration, bent neck incidence, chlorophyll fluorescence, corolla diameter fresh weigh and

solution usage) along with biochemical assays (e.g. electrolyte leakage, MDA and ABA concentrations) were evaluated in these experiments as described in Chapter 3, sections 3.1.4 and 3.3.4.

A



B

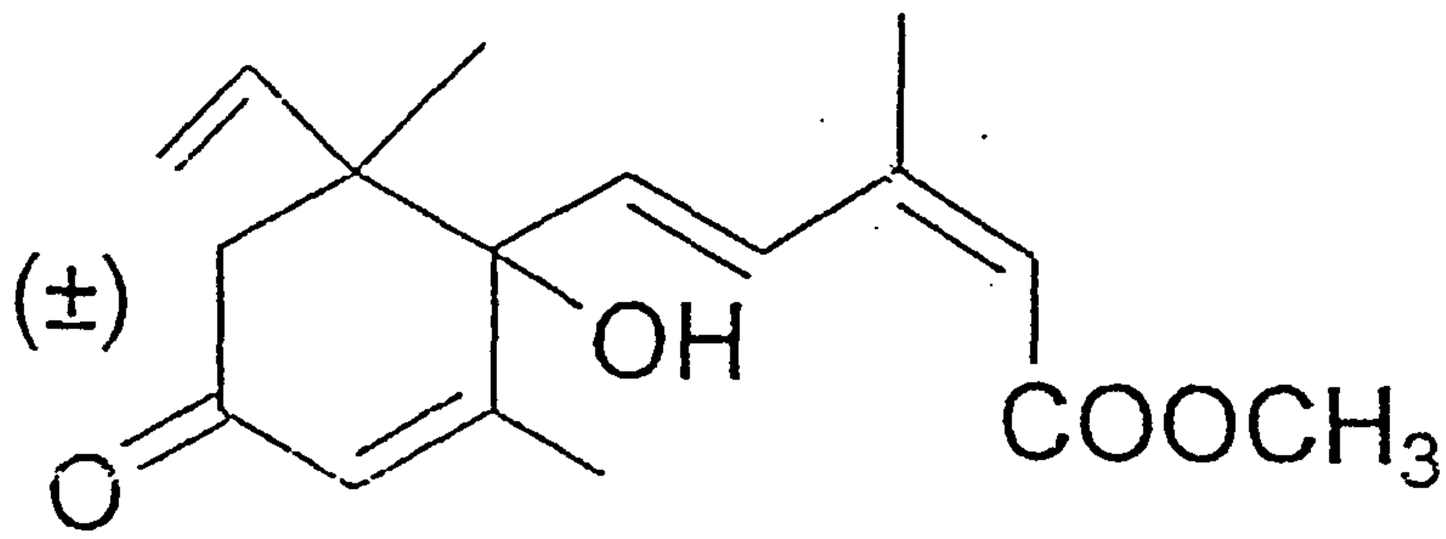


Figure 6.1: Chemical structures of (+)-ABA (A) and 8′-methylene ABA methyl (e.g. analogue PBI-365) (B).

6.2 RESULTS

6.2.1 ABA and PBI-365 effects on vase life and bent neck incidence

Pulse ABA treatment and vase solution had significant effects on vase life and bent neck of 'Akito' roses stored wet at 1°C for 10 days (Table 6.1, Appendix 6.1.1, Tables A6.1.1.1 and A6.1.1.2). Flower and foliage lives were increased by 1.8 days, when roses were pulsed with 10^{-1} M ABA before storage. This positive effect of pulse treatment on flower life (Appendix 6.1.1, Table A6.1.1.1) was more pronounced ($P < 0.05$), when vase solutions contained the analogue PBI-365. After pulsing roses with ABA, addition of ABA and PBI-365 in vase solutions increased flower life compared to control vase solution. Duncan's multiple range test showed that foliage life was significantly extended by the presence of ABA in vase solutions only when roses were not pulsed with ABA (Appendix 6.1.1, Table A6.1.1.3). However, after pulsing with 10^{-1} M ABA, vase solution containing 10^{-5} M ABA or PBI-365 did not affect foliage life. Addition of ABA in the vase solution reduced bent neck of roses either with or without pulse treatment before storage (Table 6.1). PBI-365 also reduced bent neck of roses that had not been pulsed with ABA.

Table 6.1: Vase life and bent neck of 'Akito' roses pulsed with 10^{-1} M ABA or without ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, 10^{-5} M PBI-365 and distilled water (control). Data are treatment $\bar{x}$; n = 6. ANOVA and Duncan's tests are presented in Appendix 6.1.1.

Treatments before storage	Vase solutions	Vase life (days) [a]		**Bent neck (%) [b]**
		Flower	**Foliage**	
Control	Control	6.0a	6.0 a	66.6 (4/6)
	ABA	8.0a, b	8.6b, c	33.3 (2/6)
	PBI-365	6.6a	7.3a, b	33.3 (2/6)
Column means		6.8	7.3	44.4 (2.5/6)
Pulse ABA	Control	6.6a	8.0 a, b, c	33.3 (2/6)
	ABA	10.0b	10.0c	0.0 (0/6)
	PBI-365	9.3b	9.3b, c	33.3 (2/6)
Column means		8.6	9.1	22.2 (1.3/6)

[a] Within columns, numbers followed by the same letter are not significantly different at ($P = 0.05$). [b] Bent neck data are proportions for individual treatments; n=6.

6.2.2 ABA and PBI-365 analogue effects on F_v/F_m, corolla diameter, fresh weight and solution usage changes during vase life

6.2.2.1 Relative Chlorophyll Fluorescence

Storing 'Akito' roses at 1°C for 10 days slightly reduced leaf F_v/F_m value of 0.83 (time of harvest) (Figure 6.2A, B). This slight reduction in F_v/F_m value on day 0 is similar to that reported for 'Akito' roses after storage at 1°C in Chapter 5 (section 5.2.1.2). F_v/F_m decline after storage (day 0 of vase life) was more pronounced for roses not treated with ABA compared to roses pulsed with 10^{-1} M ABA (Figure 6.2A, B). However, in both cases F_v/F_m value was maintained above 0.80 on day 0, indicating there was no physicochemical lesion on PSII activity. Thereafter, F_v/F_m value progressively declined for roses without pulse treatment. Addition of ABA or PBI-365 in vase solutions slightly maintained F_v/F_m of roses without pre-storage pulse treatment with ABA. This negligible effect of ABA and PBI-365 on F_v/F_m maintenance was not significant ($P > 0.05$) during vase life evaluation (Appendix 6.2.1, Tables A6.2.1.4 – A6.2.1.4). However, when roses were pulsed with ABA, F_v/F_m increased until day 12 followed by a decrease at the end of vase life (Figure 6.2B). In pulse treatment, leaf F_v/F_m was best maintained during vase life for roses in solutions with 10^{-5} M ABA (Appendix 6.2.1, Tables A6.2.1.5 – A6.2.1.8). The increased F_v/F_m by the presence of ABA in vase solution was significant ($P < 0.05$) the last day of vase life for pulse treatment (Appendix 6.2.1, Table A6.2.1.8). Moreover, when roses were pulsed with ABA, addition of PBI-365 in the vase solution maintained greater F_v/F_m compared to control vase solution.

6.2.2.2 Corolla diameter

ABA treatments before storage did not affect corolla diameter during vase life evaluation (Figure 6.2C, D). However, vase solution containing ABA or PBI-365 tended to increase corolla diameter for roses pulsed with or without ABA. In non-pulse treatment (Figure 6.2C), flowers in solutions with ABA had greatest corolla diameter on days 4 and 8 followed by those in solutions with PBI-365. Corolla diameter for these flowers reached the maximum diameter of *ca.* 90 and 80 mm around day 4 for treatments of ABA and PBI-365, respectively. Increased corolla

diameter for treatments of ABA and PBI-365 was significant on day 4 compared to control solutions (Appendix 6.2.2, Tables A6.2.2.1 – A6.2.2.5). When roses were pulsed with ABA before storage at 1°C (Figure 6.2C, Appendix 6.2.2, Tables A6.2.2.6 – A6.2.2.10), the presence of ABA or PBI-365 in the vase solutions tended to slightly, but non-significantly ($P > 0.05$), increase corolla diameter compared to control solutions.

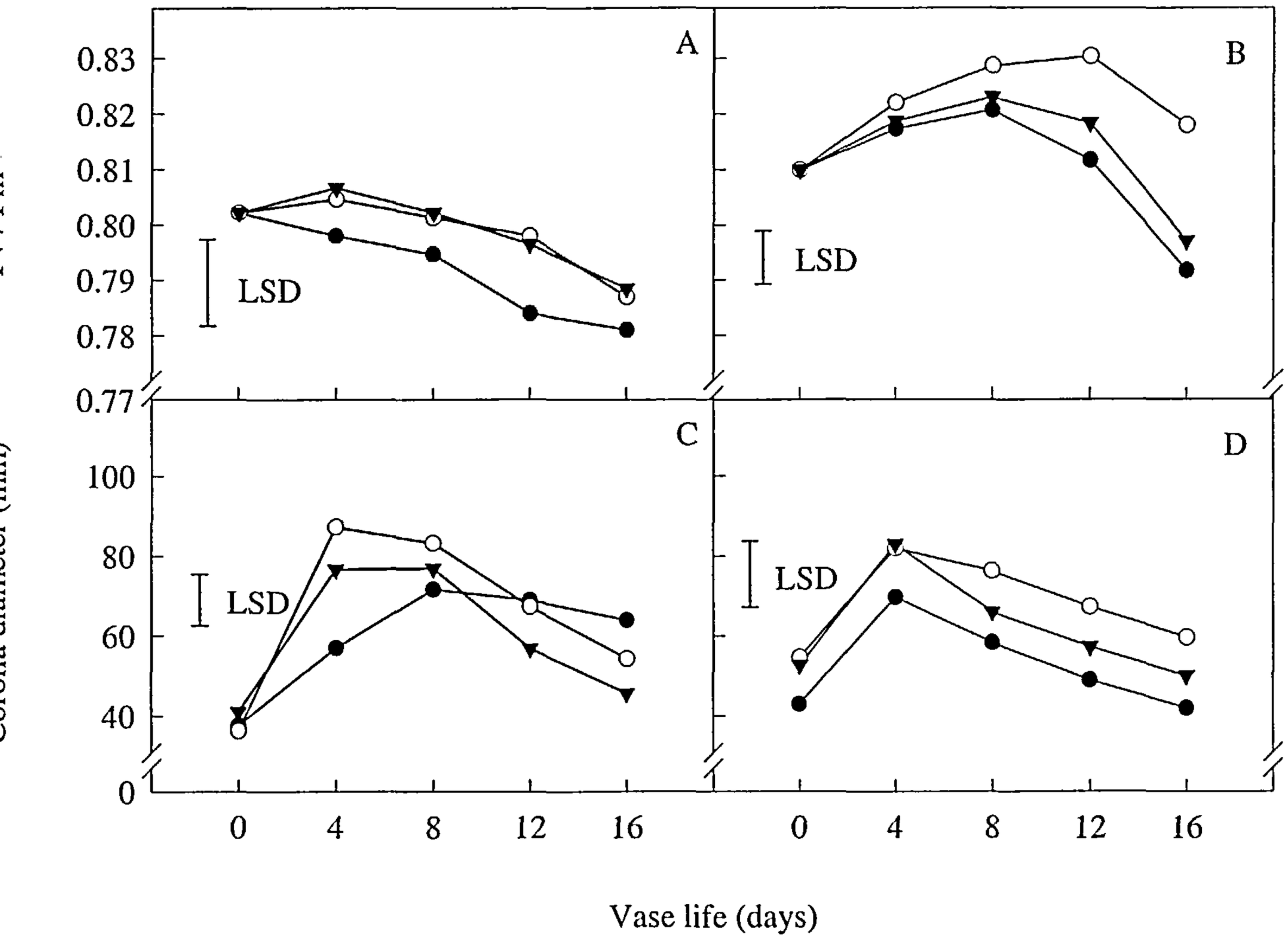


Figure 6.2: Changes in corolla diameter (A, B) and F_v/F_m (C, D) during vase life for 'Akito' roses pulsed with 10^{-1} M ABA (B, D) or without ABA (control) (A, C) and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA (○), 10^{-5} M PBI-365 (▼) and distilled water (control) (●). Data are independent treatment means; n = 6. Vertical bars show ± L.S.D. (P = 0.05). ANOVA tables are presented in Appendices 6.2.1 and 6.2.2.

6.2.2.3 Relative fresh weight

Fresh weight generally increased during the first two days of vase life (Figure 6.3A, B, Appendix 6.2.3, Tables A6.2.3.1 – A6.2.3.16). The increase of fresh weight in cut roses usually takes place the first days of vase life, but afterwards fresh weight decreased, indicating reduction of water uptake (Halevy and Mayak, 1981). Fresh weight changes during vase life were unaffected by pulse ABA treatment (Figure 6.3A, B). On the other hand, vase solutions containing 10^{-5} M ABA maintained higher fresh weight compared to the solutions in the absence of ABA, especially after day 6. Fresh weight was also maintained by addition of 10^{-5} M PBI-365, but to lesser degrees. Both ABA and PBI-365 had greater capacity in maintaining fresh mass when added to vase solution after pulse treatment (Figure 6.3B, Appendix 6.2.3, Tables A6.2.3.9 – A6.2.3.16).

6.2.2.4 Vase solution usage

The rate of solution usage by flowers in solutions without ABA or PBI-365 was generally highest during vase life (Figure 6.3C, D). Reduced solution usage with ABA or PBI-365 compared to control solutions was recorded from the time that flowers were placed into vases until the last day of the experiment. For pulse treatment (Figure 6.3D, Appendix 6.2.4, Tables A6.2.4.1 – A6.2.4.8), reduction of solution usage brought about by ABA or PBI-365 was significant from 3 to 9 day compared to control solutions. When roses were not pulsed with ABA (Figure 6.3C, Appendix 6.2.4, Tables A6.2.4.9 – A6.2.4.16), they had lowest solution usage in solutions with PBI-365. Both ABA and PBI-365 had greater capacity in reducing solution usage by flowers when added to vase solution after pulse treatment (Figure 6.3D, Appendix 6.2.4, Tables A6.2.4.1 – A6.2.4.8). End of vase life was associated with much reduced solution usage by flowers in all solutions.

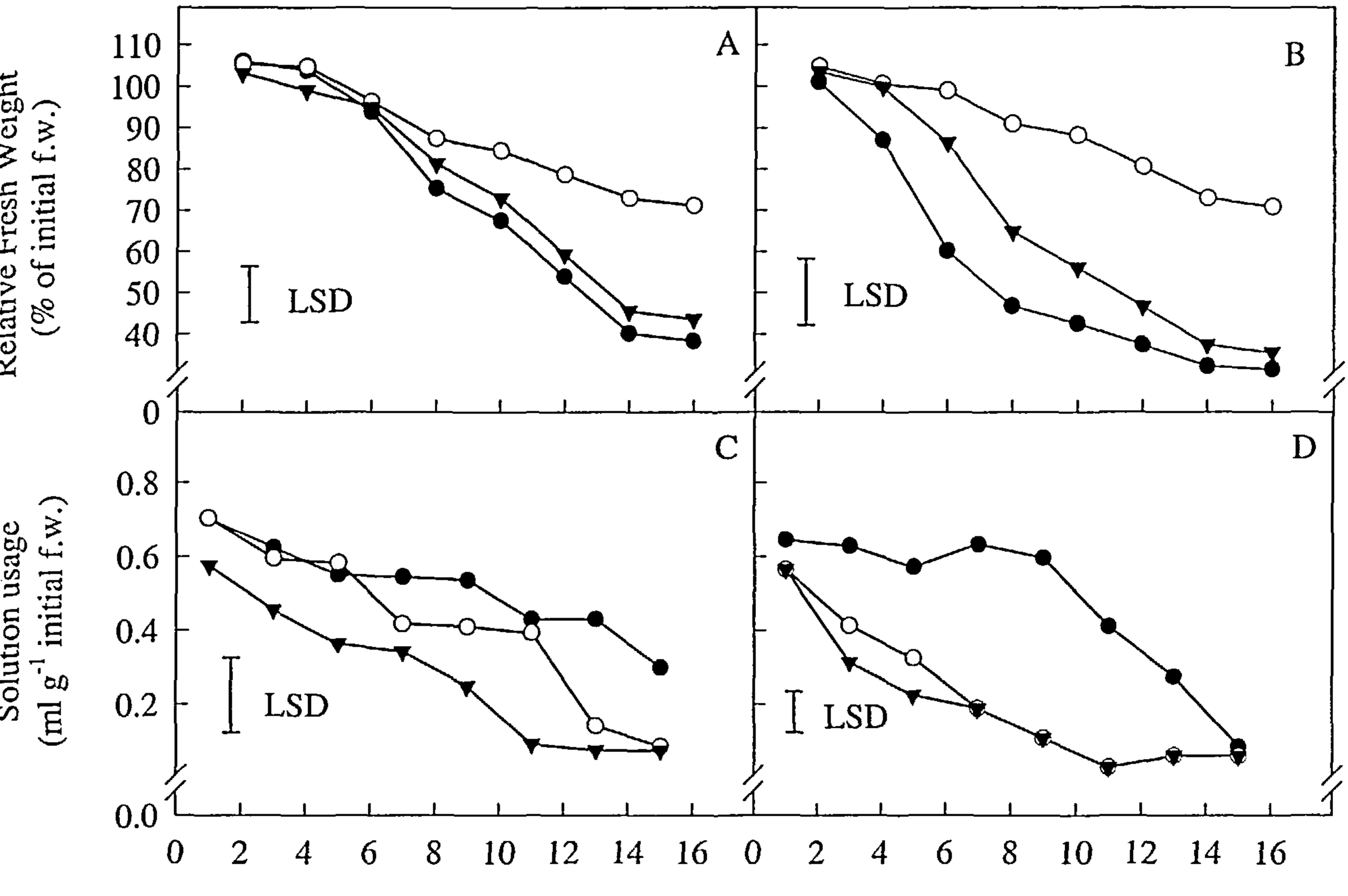


Figure 6.3: Fresh weight and solution usage changes during vase life by 'Akito' roses pulsed with 10^{-1} M ABA (B) or without ABA (A) and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA (○), 10^{-5} M PBI-365 (▼) and distilled water (●). Data are independent treatment means; n = 6. Vertical bars show ± L.S.D. (P = 0.05). ANOVA tables are presented in Appendix 6.2.3 and 6.2.4.

6.2.3 ABA and PBI-365 analogue effects on biochemical assays

6.2.3.1 Electrolyte leakage

Neither pulse treatments nor vase solutions had significant effects on electrolyte leakage in petals of roses at the middle of vase life (Table 6.2, Appendix 6.3.1, Table A6.3.1.1). Electrolyte leakage was lower for flowers in solutions with ABA or PBI-365 compared to control solutions; however, this difference was not

significant (P > 0.05). Pulsing roses with 10^{-1} M ABA reduced electrolyte leakage in leaves (Table 6.2), although this effect was not significant (Appendix 6.3.1, Table A6.3.1.2). The presence of ABA in the vase solutions significantly reduced electrolyte leakage in leaves either with or without pulse treatment (Table 6.2, Appendix 6.3.1, Table A6.3.1.3). Similarly, addition of PBI-365 significantly (P < 0.05) reduced electrolyte leakage in leaves after pulsing with ABA.

Increasing electrolyte leakage in petals and leaves was positively correlated with decline in both flower (r^2 = 0.61) and foliage (r^2 = 0.79) lives, respectively (Figures 6.4A and 6.5A, Appendix 6.3.3, Table A6.3.3.5). Because of the association between electrolyte leakage and foliage life, electrolyte leakage may provide an index of foliage life.

Table 6.2: Electrolyte leakage (%) in petals and leaves of 'Akito' roses pulsed with 10^{-1} M ABA or without ABA (control) and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, 10^{-5} M PBI-365 and distilled water (control). Data are individual treatment means; n = 6. ANOVA and Duncan's tests are presented in Appendix 6.3.1.

Treatments before storage	**Vase solutions**	Electrolyte leakage (%) [a]	
		Petals [b]	**Leaves**
Control	Control	11.9a	54.4a
	ABA	5.4a	19.8b, c
	PBI-365	5.7a	31.2a, c
Column means		7.6	35.1
Pulse ABA	Control	13.5a	46.8a, b
	ABA	3.8a	11.9c
	PBI-365	4.0a	13.8c
Column means		7.1	24.1

[a] Electrolyte leakage was measured on day 10 of vase life. [b] Within columns, numbers followed by the same letter are not significantly different at (P = 0.05).

6.2.3.2 Malondialdehyde (MDA) content in petals and leaves

MDA content in leaves was 3-fold greater than that in petals of roses at the middle of vase life (Table 6.3). Pulse ABA treatment significantly (P < 0.05) decreased MDA content in leaves by 1.7-fold (Appendix 6.3.2, Table A6.3.2.3).

Provision of ABA or PBI-365 in vase solution decreased non-significantly MDA content in petals and leaves of roses that had not been pulsed with ABA. However, when roses were pulsed before storage, MDA content in petals and leaves was significantly reduced by 1.6- and 3-fold in solutions with ABA and PBI-365, respectively (Appendix 6.3.2, Tables A6.3.2.2 and A6.3.2.4).

Table 6.3: Malondialdehyde (MDA; mM^{-1} cm^{-1}) content in petals and leaves of 'Akito' roses pulsed with 10^{-1} M ABA or without ABA (control) and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, 10^{-5} M PBI-365 and distilled water (control). Data are individual treatment means; n = 6. ANOVA and Duncan's tests are presented in Appendix 6.3.2.

Treatments before storage	**Vase solutions**	MDA (mM^{-1} cm^{-1}) [a]	
		Petals [b]	**Leaves**
Control	Control	13.6a, c	55.2a
	ABA	10.3a, b	42.1a, b
	PBI-365	10.7a, b	45.3a, b
Column means		11.5	47.5
Pulse ABA	Control	15.2c	36.6b
	ABA	9.5b	34.2b
	PBI-365	12.9a, c	12.2c
Column means		12.5	27.5

[a] MDA concentration was measured on day 10 of vase life. [b] Within columns, numbers followed by the same letter are not significantly different at (P = 0.05).

6.2.3.3 ABA content in petals and leaves

ABA concentrations in leaves were 3.2-fold greater than those in petals (Table 6.4). ABA concentration in petals and leaves at the middle of vase life (day 10) was significantly affected by pulse ABA treatments before storage and vase solutions after storage (Table 6.4, Appendix 6.3.3, Tables A6.3.3.1 and 6.3.3.3). Exogenous ABA supplied in the water of roses before storage for 24 hours increased ABA content in petals and leaves 3.2- and 2-fold, respectively (Table 6.4, Appendix 6.3.3, Tables A6.3.3.2 and A6.3.3.4). Significant increases in ABA contents by pulse ABA treatment were evident for all vase solutions. Increased ABA concentration was

detected in petals and leaves, when rose stems were placed in vase solution containing ABA followed by those placed in solutions with PBI-365.

Longer flower and foliage lives were linearly correlated with increasing ABA content in petals (r^2 = 0.61) and leaves (r^2 = 0.75), respectively (Figures 6.4B and 6.5B, Appendix 6.3.3, Table A6.3.3.5). These associations indicate a strong relationship between ABA levels and vase life duration in roses.

Table 6.4: ABA content (ng g^{-1} f.w.) in petals and leaves of 'Akito' roses pulsed with 10^{-1} M ABA or without ABA (control) and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, 10^{-5} M PBI-365 and distilled water (control). Data are individual treatment means; n = 3. ANOVA and Duncan's tests are presented in Appendix 6.3.3.

Treatments before storage	Vase solutions	ABA content (ng g^{-1} f.w.) [a]	
		Petals [b]	**Leaves**
Control	Control	89.4a	703.9a
	ABA	117.2a	1097.9b
	PBI-365	97.4a	729.7a
Column means		101.3	843.8
Pulse ABA	Control	225.3b	1263.1b, c
	ABA	508.7c	2370.5d
	PBI-365	235.7b	1491.4c
Column means		323.2	1708.3

[a] MDA concentration was measured on day 10 of vase life. [b] Within columns, numbers followed by the same letter are not significantly different at (P = 0.05).

6.2.3.4 Glasshouse environmental conditions

In the present study, two successive vase life experiments were carried from February 2004 to April 2004 (Chapter 3, section 3.4.1). Mean day and night temperatures progressively increased and, as a result, RH declined from February to April (Figure 6.6A, B). In association with greater temperatures, PFD increased from February to April reaching a maximum of *ca.* 1800 µmol m^{-2} sec^{-1} (Figure 6.6C). The same period in 2003 (Chapter 4, section 4.2.2.1), roses were grown in lower temperature and PFD. For example, mean day temperatures in March 2003 and 2004

were *ca.* 20 and 25°C, respectively. Similarly, mean PFD in March 2003 and 2004 was ca. 1350 and 1500, respectively. Mean day and night RH was greater in 2003 than in 2004.

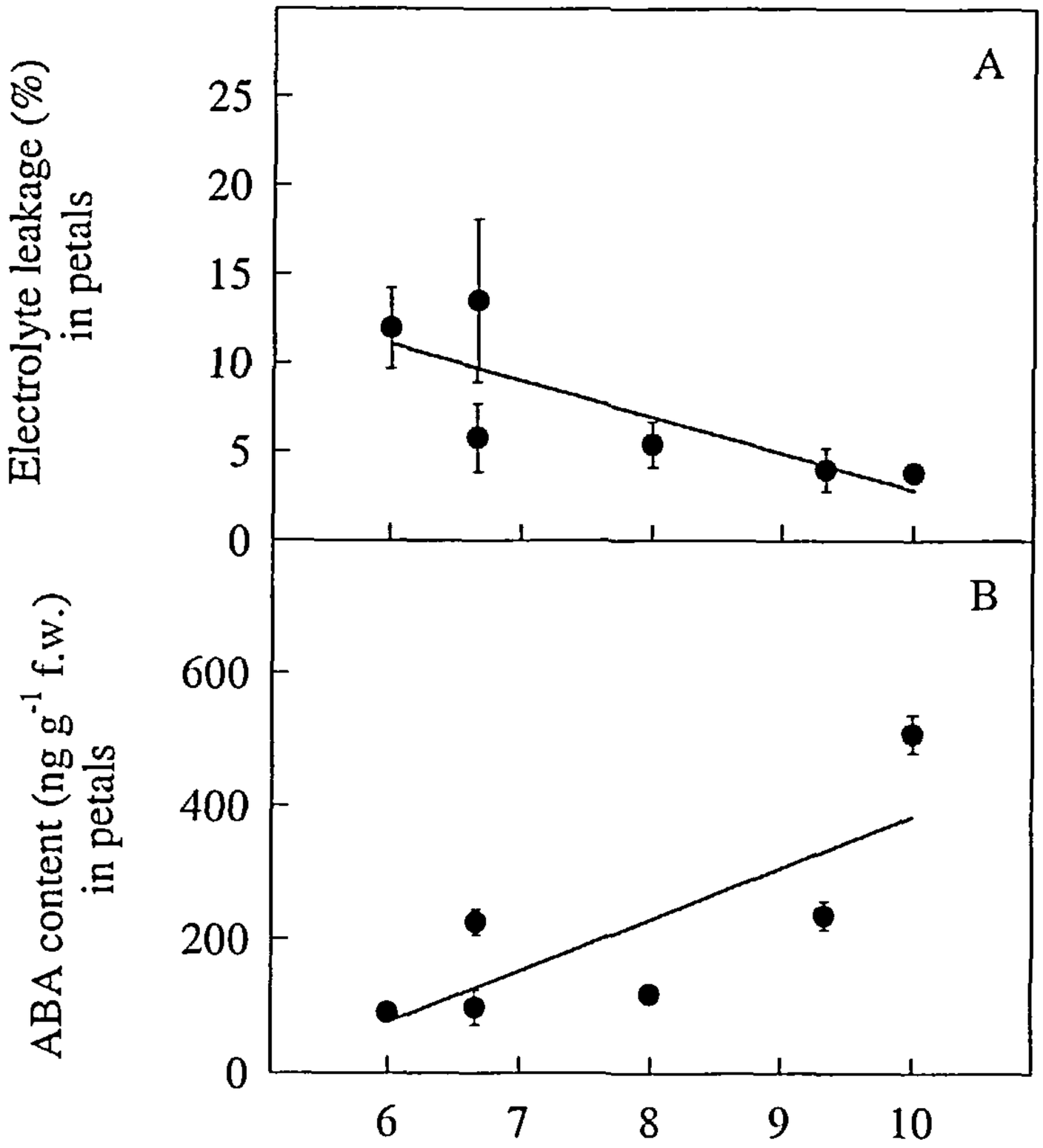


Figure 6.4: Simple linear regression analysis (y = a.x +/- b) between flower life and electrolyte leakage (%) (A) or ABA content (ng g^{-1} f.w.) (B) in petals of 'Akito' roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water. Data are $\bar{x}$; n = 6 for electrolyte leakage, n = 3 for ABA. Vertical bars on scatter plot show ± S.E. (n = 6 for electrolyte leakage, n = 3 for ABA) for each flower life duration. Regression parameters are presented in Appendix 6.3.3, Table A6.3.3.5.

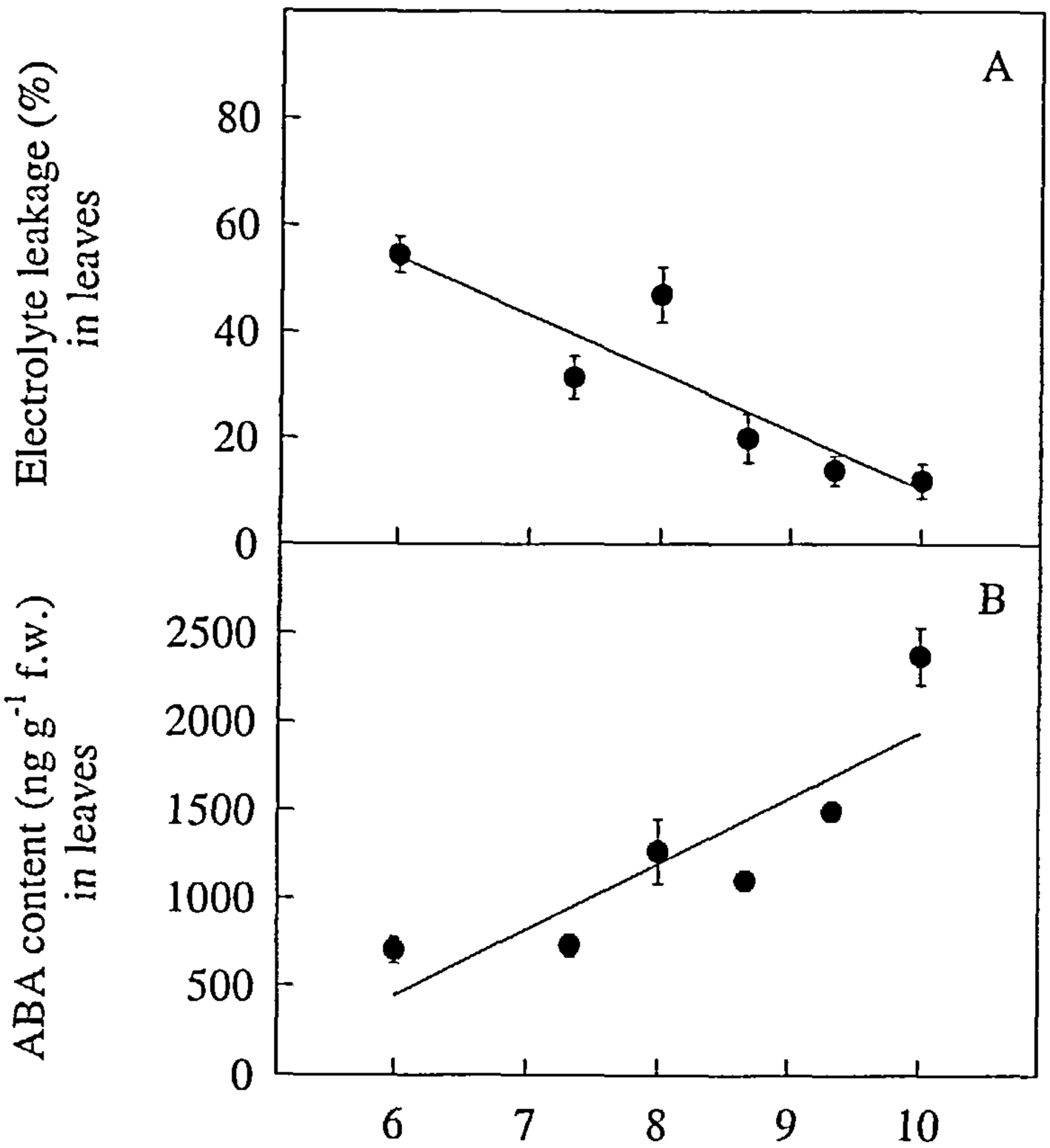


Figure 6.5: Simple linear regression analysis (y = a.x +/- b) between foliage life and electrolyte leakage (%) (A) or ABA content (ng g^{-1} f.w.) (B) in leaves of 'Akito' roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water. Data are $\bar{x}$; n = 6 for electrolyte leakage, n = 3 for ABA. Vertical bars on scatter plot show ± S.E. (n = 6 for electrolyte leakage, n = 3 for ABA) for each foliage life duration. Regression parameters are presented in Appendix 6.3.3, Table A6.3.3.5.

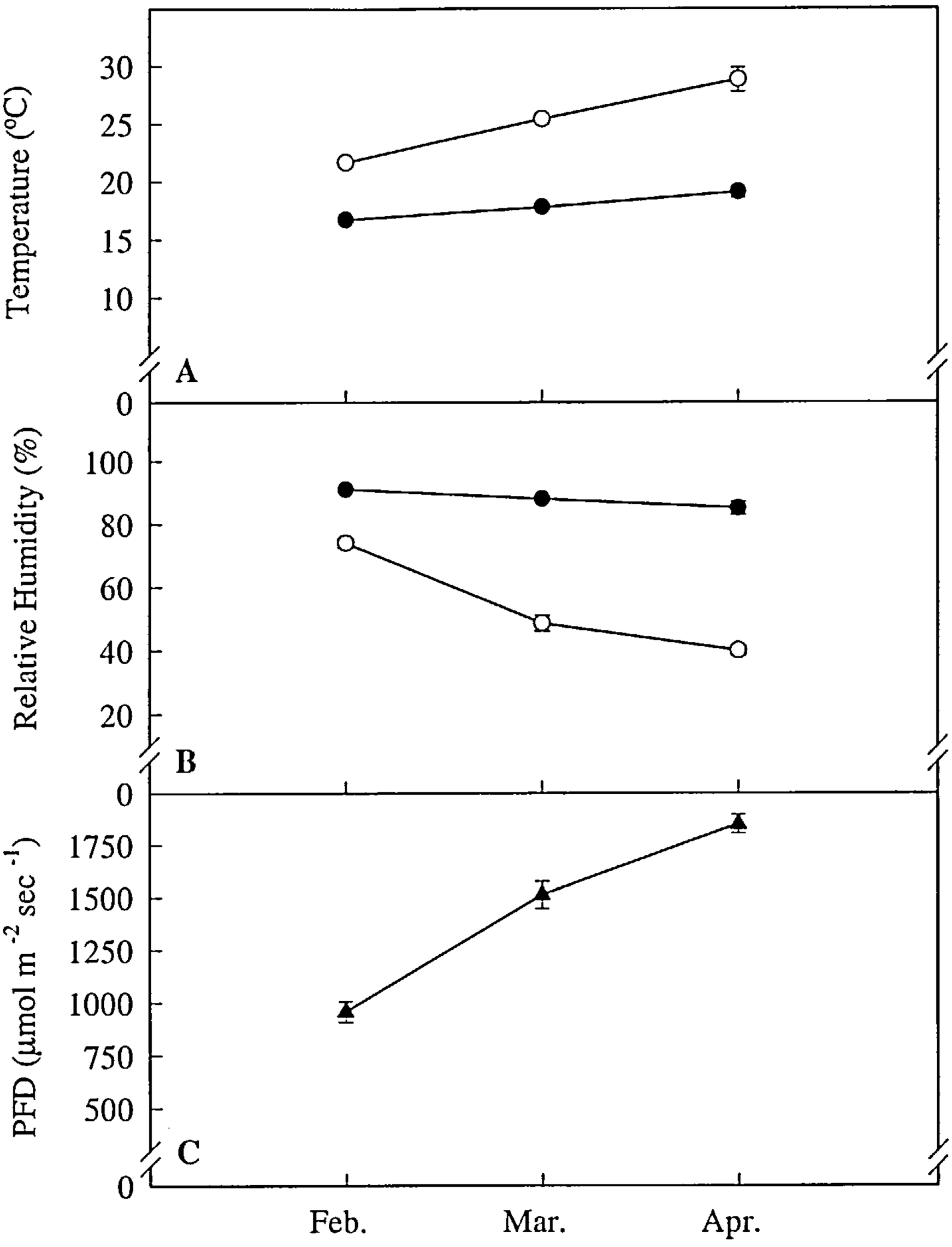


Figure 6.6: Changes in (A) temperatures (°C) and (B) RH (%) during the day (○) and night (●), and (C) Photon Flux Density (PFD) (µmol m^{-2} s^{-1}) in the glasshouse environments throughout the growing period (from February 2004 to April 2004). Data are means of 12 recordings per day; *n* = number of days each month. Vertical bars show ± SE for each month (*n* = number of days each month).

6.3 DISCUSSION

Direct evidence for a protective function of ABA against LTI was achieved in this study by application of the hormone itself or with the ABA synthetic analogue PBI-365. Provision of 10^{-5} M ABA or PBI-365 in the vase solution generally increased flower and foliage lives of roses. Increased vase life with ABA or PBI-365 was more pronounced, when roses had been pulsed with 10^{-1} M ABA, indicating a synergic effect of pulse treatments and vase solutions. Moreover, pulse ABA treatment along with addition of ABA in vase solution reduced bent neck incidence. In the present study, ABA and the analogue PBI-365 apparently caused stomatal closure, since the transpiration rate was reduced during vase life (Trejo *et al.*, 1993; Halevy *et al.*, 1974). Induction of stomatal closure is also supported by the fact that increased ABA levels were detected in leaves after exogenous application with ABA before storage (pulse treatment) or during vase life (vase solution). These data are in agreement with previous work on cut 'Baccara' roses, which indicated lower water uptake using ABA or PBI-365 (Pompodakis and Joyce, 2003). However, in cut 'Baccara' roses, the reduced solution usage by ABA or PBI-365 did not translate into either flower or foliage vase life extension. ABA also maintained greater fresh mass of cut roses throughout vase life, as a result of possible stomatal closure and decreased transpiration rates. On the other hand, when flowers were not treated with ABA or PBI-365, they had shorter vase lives, greater water uptake and less fresh weight maintenance, indicating increased water loss through stomata. Increased water loss and inability of plants to close their stomata at low temperature, without exogenous application with ABA, has been previously reported for chilling sensitive species, such as *Phaseolus vulgaris* L. and *Pisum sativum* L. (Wilson, 1976; Eamus *et al.*, 1983). However, this finding was reported pre-harvest and may have no implications on postharvest studies.

Increased ABA content in petals and leaves was associated with longer flower and foliage lives, respectively. The relationship between foliage life and ABA levels in leaves ($r^2 = 0.75$) was stronger than that between flower life and ABA content in petals ($r^2 = 0.61$), indicating a greater influence of ABA on leaf water stress. This could simply be attributed to ABA-enhanced stomatal closure and, thereby, reducing water loss (Davies *et al.*, 2002). However, in 'stomata-less' systems, such as rose

petals (Stubbs and Francis, 1971; Mayak and Halevy, 1974; van Doorn and Vojinovic, 1996), ABA is less effective in extending vase life. ABA content in petals of 'Akito' roses without pulse treatment was not affected significantly by the presence of ABA in the vase solution. Similarly, Muller *et al.* (1999) reported that ABA treatment did not change significantly ABA content in petals of 'Vanilla' roses. However, when 'Akito' roses were pulsed with ABA before storage, addition of ABA in the vase solution significantly increased ABA content in petals. This significant increase in ABA content might be an interactive effect of low temperature storage and ABA pulse treatment.

Storing 'Akito' roses at 1°C for 10 days did not result in F_v/F_m decline. Similarly, F_v/F_m value on day 0 was not affected by storage temperature in Chapter 5. These similar results of F_v/F_m values after storage at 1°C may be attributable to the same growing conditions (e.g. temperature, RH, PFD) of current experiments and those of Chapter 5. In Chapter 4, however, leaf F_v/F_m declined for autumn- and winter-grown roses after storage at 1°C, indicating LTI effects on PSII activity. The lower F_v/F_m of 'Akito' roses in first year experiments (Chapter 4) is apparently due to lower PFD and/or growing temperature compared to current experiments and Chapter 5. The sharp fall of F_v/F_m for roses grown in winter (lower PFD and temperature) was due to an interactive effect of low temperature storage and growing season (Chapter 4, section 4.3.1 and Chapter 5, section 5.3.1). In the present study, when roses were treated with ABA before storage, they recovered slightly their quantum yield of PS II (F_v/F_m) during vase life evaluation. F_v/F_m recovery was greater for roses in solutions with ABA followed by those in solutions with PBI-365. F_v/F_m recovery was apparently due to induction of stomatal closure by ABA or PBI-365 and the subsequent decline in transpiration rate resulting in less water stress (Pardossi *et al.*, 1992). Similarly, Aroca *et al.* (2001) found that intact leaves of maize were capable to recover F_v/F_m 5 days after chilling at 5°C.

Pulse ABA treatment and/or vase solutions with ABA tended to decrease electrolyte leakage in petals (non-significant effects). Similarly, pulsing roses with ABA before storage at 1°C reduced the rate of electrolyte leakage in leaves by 1.5-fold. Moreover, after pulsing roses with ABA, low temperature-induced electrolyte leakage in leaves significantly ($P < 0.05$) decreased by 4- and 3.4-fold in solutions with ABA and PBI-365, respectively. Similar reductions in electrolyte leakage caused by ABA have been reported in cucumber (Flores *et al.*, 1988). The reduced

electrolyte leakage in cucumber, which was caused by exogenous application of 10^{-4} M ABA prior to chilling at 1.5°C, was related to changes in the composition and activity of membrane system (e.g. stabilisation of phospholipids). In roses, pulse ABA treatment along with the presence of PBI-365 in vase solution significantly reduced MDA content in leaves and, as a result, the degree of lipid peroxidation. In view of this, ABA and/or PBI-365 might be effective in stabilising phospholipids and preventing chilling-induced oxidation of lipids.

6.4 CONCLUSION

Use of ABA and/or synthetic ABA analogues after low temperature storage is an interesting prospect for reducing water loss and ameliorating LTI in roses. The present replicate experiments investigated the potential to improve vase life parameters of 'Akito' roses by application of ABA (pulse and vase solution) and the synthetic ABA analogue PBI-365 (vase solution). Vase solutions with ABA or PBI-365 extended both flower and foliage lives and reduced bent neck, especially when combined with pre-storage (pulse) treatment with ABA. These positive effects of ABA and PBI-365 were apparently due to induction of stomatal closure, since solution usage was reduced during vase life evaluation. Similar promising results were reported using PBI-365 as vase solution ingredient for 'Baccara' roses (Pompodakis and Joyce 2003). Thus, increased ABA content in petals and leaves was correlated with longer flower and foliage lives, respectively.

There was evidence for amelioration of LTI in roses by using ABA and PBI-365. Although there was no considerable reduction in F_v/F_m after storage, both ABA and PBI-365 helped to recover F_v/F_m value during vase life. Electrolyte leakage in petals and leaves of roses stored at 1°C was reduced by pulse and vase solutions with ABA. Additionally, lipid peroxidation in leaves was inhibited when roses had been pulsed with ABA before storage and then were put in vase solutions containing PBI-365. However, it remained unclear whether ABA and PBI-365 reduced LTI directly by inducing stomatal closure or other mechanisms (e.g. stabilisation of phospholipids, inhibition of phospholipid hydrolysis, enhancement of antioxidant enzymes), are involved in the mode of ABA and PBI-365 action.

CHAPTER 7

OVERALL CONCLUSIONS AND RECOMMENDATIONS FOR FUTURE RESEARCH

Suitable postharvest storage and transport procedures are required by cut rose industry in Mediterranean to develop and promote exports (G. Chalkiadakis and M. Fthenakis pers. comm., 2001). This thesis is one of the first studies on post-storage characteristics of roses grown in Mediterranean all year round. The work presented has demonstrated the potential to improve vase life of roses after cold storage using the plant growth regulator ABA and a novel ABA analogue (e.g. PBI-365). Both compounds were used as protective agents against Low Temperature Injury (LTI). Chlorophyll fluorescence and the sensitivity of roses to LTI after storage were related to seasonal variation during the year. Improvement of vase life characteristics after cold storage was achieved to varying degrees by both ABA and PBI-365. However, considerable research is required to investigate more practical implications by the use of ABA and analogue.

7.1 EFFECT OF SEASONAL VARIATION ON VASE LIFE OF ROSES

Pre-harvest environmental conditions were investigated during the year with a view to discerning a relationship between seasonal variations and vase life parameters of roses. Vase life duration of both 'First Red' and 'Akito' roses, reported in the present study, followed a seasonal pattern in 2003-04, similar to that found for 'Orange Unique' and 'Diva' roses by Slootweg *et al.* (2001). Reduction in vase life was recorded in autumn- (September, October and November) and winter-months (December, January and February) as compared to spring (March, April and May) and summer (June, July and August). Environmental conditions through out the year (e.g. temperature, Photon Flux Density, PFD and RH) monitored inside and outside two rose glasshouses in Crete were correlated with vase life duration. The maximum potential quantum yield of PS II (F_v/F_m) monitored at the beginning of vase life for non-stored control roses did not vary between growing seasons. Thus, this parameter was not capable of assessing possible pre-harvest variations in rose condition during the year. However, when roses were stored for 10 days, F_v/F_m measured on day 0 of

vase life was strongly affected by growing season and storage temperature. Greatest F_v/F_m losses were recorded for winter-grown roses after storage at 1°C suggesting synergistic effects of growing conditions and storage temperature on LTI of roses. The negative F_v/F_m responses of winter-grown roses to low temperature storage were correlated with low PFD during cultivation. Thus, the susceptibility of winter-grown roses to LTI reported here has been attributed to reduced carbohydrate pools due to lower photosynthesis (King *et al.*, 1988). In contrast, the second year growing temperature and PFD were consistently higher during cultivation resulting in less F_v/F_m losses for 'Akito' roses after storage.

In addition, seasonally dependent fluctuations in environmental conditions seemingly affected vase life quality. b^* value measured on petals of 'First Red' roses decreased in autumn and winter, indicating greater petal blueing at the middle of vase life. There is strong evidence in the literature that low light intensities and/or UV-radiation in autumn and winter months accelerate petal blueing through inhibition of anthocyanin synthesis (Drumm-Herrel, 1984; Chalker-Scott, 1999). However, there were no correlative effects of environmental conditions on b^* value for 'First Red' roses to support these findings. Additionally, considerable reduction in flower opening of 'Akito' roses was recorded in autumn and winter using a subjective rating scale with five developmental stages. The strong association between reduced flower opening and PFD decline from September to February suggested photosynthetic activity in glasshouse to be a limiting factor for flower opening during vase life. Similar results in roses were attributed to lack of carbohydrate pools in the corolla (Marissen, 2001). More research is needed to improve knowledge on physiological responses of different flower parts (receptacle, petals, stamens, styles) in relation to seasonal variation in environmental conditions.

Bent neck symptom in roses was strongly affected by genotype. Bent neck incidence of 'Akito' was greater compared to 'First Red' roses throughout the year. According to the results presented here, the great sensitivity of some cultivars, such as 'Akito' roses, to bent neck disorder may be explained by: 1) increase of water losses by leaves during the first 48 hours after harvest, which may have accelerated water stress, 2) excessive water uptake throughout vase life evaluation, and 3) lack of adequate lignified xylem elements within vascular bundles in the peduncle region. Enhancement of bent neck symptom under water stress conditions was also reported by Burdett (1970) and by Zieslin *et al.* (1978) for 'Forever Yours', 'Cara Mia',

Jaqueline', 'Samantha' and 'Town Crier' roses. Extent of lignification in peduncles was reduced by growing roses from September 2003 to February 2004, suggesting a possible role of seasonality on lignin synthesis and/or accumulation within plant cells. Therefore, it is essential to continue investigations on the relationship between variation in lignin concentration and seasonal changes during the year using more precise methods for lignin quantification. The activity of certain precursors/promoters (e.g. phenylalanine ammonia lyase; PAL) of lignin synthesis in minimizing bent neck incidence in cut roses also warrants investigation.

Vase solution microflora (e.g. bacteria, fungi and yeasts) is the major cause of vascular occlusions (van Doorn, 1997) for roses. The degree of microbial growth represented by vase solution turbidity was tested in relation to seasonal variation in growing environment. Greater solution turbidity was recorded at the end of vase life for roses grown from March (spring) to August 2002 (summer) than over the rest of the year. Increased solution turbidity during spring and summer was attributed to greater microbial growth and deposition of materials from the cut stem to vase solution. Metabolites such as carbohydrates, amino acids and amides (Halevy *et al.*, 1974) from flower stem and microbial degradation products may leach into the vase solution (van Doorn, 1997). Further study at the cellular level is warranted to investigate vascular occlusions in relation to environmental conditions in glasshouse.

7.2 EFFECT OF STORAGE TEMPERATURE ON VASE LIFE OF ROSES

The vase life parameters were evaluated after storing cut 'First Red' and 'Akito' roses wet at low temperature (10, 5, 1°C) for 10 days. Increasing storage temperature of 'First Red' roses from 1 to 10°C was associated with vase life reductions, increased petal blueing and lesser capacity to maintain fresh weight during vase life. These reproducible findings for 'First Red' roses were evidently due to enhanced senescence during storage at relatively higher storage temperature as reported by Faragher and Mayak (1984) and Faragher *et al.* (1984, 1986) for 'Mercedes' roses. In 'Akito' roses, however, storage temperature did not exert significant effects on flower or foliage longevity in first year experiments (Chapter 4, section 4.2.1.1). By contrast, the second year (Chapter 5, section 5.2.1.1)

demonstrated greater flower and foliage life losses for 'Akito' roses stored at 1°C rather than at 5°C, suggesting possible enhancement of LTI.

Induction of LTI in both cultivars during low temperature storage was strongly supported by decline in F_v/F_m at the beginning of vase life. Storage of roses at 1°C compared to 5°C and more so at 10°C induced a considerable reduction in F_v/F_m of roses grown in winter and, to a lesser extent, in autumn. The fall of F_v/F_m was greater for 'Akito' roses compared to 'First Red' indicating that the sensitivity of roses to LTI is strongly affected by genotype. Reduction in F_v/F_m at low storage temperature reflects a physicochemically-based reduction in light energy utilization by chloroplasts caused by LTI. Membranes of low temperature or chilling sensitive cells undergo alterations in biophysical properties that can alter functionality. An effect of CI is thylakoid unstacking in chilling sensitive plant tissues resulting in the rapid chloroplast deterioration (Marangoni *et al.*, 1996). The sharp decline in F_v/F_m after storage at 1°C did not, however, translate into a correspondingly sharp reduction in vase life. Although there was significant correlation ($P \leq 0.01$) between F_v/F_m on day 0 and vase life, this relationship was very weak; $r^2 = 0.43$ and 0.44 for 'First Red' and 'Akito' roses, respectively. Thus, while the CF index F_v/F_m might be used to predict a LTI related physicochemical lesion in PSII of rose leaf chloroplasts, it is not a practically useful predictor of rose vase life. This apparent discrepancy presumably arises because rose vase life is determined more by temperature effects on rates of deterioration (e.g. petal fading, leaf desiccation) than by LTI.

Biochemical assays were performed during vase life evaluation to determine physiological responses of cut roses to low temperature storage. Storing 'Akito' roses at 1°C increased electrolyte leakage and lipid peroxidation (e.g. MDA content) in leaves 1.4- and 1.6-fold, respectively. Such biochemical lesions during and/or after low temperature exposure have been previously attributed to modifications of membrane permeability and alterations of enzyme activities (Murata, 1989). However, storage temperature did not alter electrolyte leakage or the degree of lipid peroxidation in 'First Red' roses. This finding further supports that the development of LTI in roses is related to genotype sensitivity. In view of these results, further investigations of LTI using additional rose cultivars are needed. Also, research at the molecular level is warranted to help interpret the LTI mode of action in roses.

Vardi and Mayak (1989) and Orlandini (1991) showed that the amount of ABA can increase substantially as a function of developed water stress. Thus, the possible water stress during storage may enhance ABA biosynthesis in roses. Concomitant to this, ABA accumulation increased *ca.* 1.53- and 1.59-fold in petals and leaves of 'Akito' roses, respectively, after low temperature storage in this present study. In tomato and other chilling-sensitive species, ABA biosynthesis is caused directly by low temperature without enhancement of water stress (Starck *et al.*, 1998). Therefore, more research is needed to investigate whether the increased ABA concentration in roses reported here is related to water stress during storage or whether other mechanisms are involved.

7.3 EFFECT OF PRE- AND POST-STORAGE APPLICATION OF ABSCISIC ACID IN IMPROVING VASE LIFE OF ROSES

The capacity of the plant growth regulator abscisic acid (ABA) in improving vase life parameters after low temperature storage was tested for cut roses. There was strong evidence for extension of vase life and inhibition of bent neck for roses pulsed before storage with ABA. Also, provision of ABA in vase solution of 'Akito' roses after storage at 1°C was markedly effective in extending vase life, reducing bent neck and maintaining fresh weight during vase life evaluation. These findings agree with previous work on roses (Kohl and Rundle, 1972; Halevy *et al.*, 1974; Pompodakis and Joyce, 2003) and Geraldton waxflower (Joyce *et al.*, 1996) which showed that exogenous ABA treatment extended longevity by reducing water loss during vase life. Moreover, increased ABA content in petals and leaves of 'Akito' was associated with greater flower and foliage lives, respectively. It is possible that the applied ABA may reach the guard cells on leaves and function as promoter of stomata closure (Wilkinson and Davies, 2002). Enhancement of stomatal closure was also supported by the reduced solution usage of roses in vase solutions containing 10^{-5} M ABA. ABA applied as pulse treatment before storage or incorporated into vase solution after storage increased ABA levels in petals and leaves providing evidence for hormone accumulation and/or enhancement of ABA biosynthesis. Further research is needed to investigate whether the increased ABA accumulation reported here is due to enhanced ABA biosynthesis. Therefore, assays using exogenous radio-labelled ABA

(Gowing *et al.*, 1993) may help in better understanding the mode of ABA action in roses.

Spraying ABA onto the leaves of 'First Red' roses prevented F_v/F_m decline brought about by storage at 1°C. Furthermore, F_v/F_m was better maintained during vase life for 'Akito' roses treated with ABA compared to control untreated flowers. These findings may suggest a protective function of ABA against LTI in cut roses possibly similar to those summarised by Janowiak *et al.* (2002) for other chilling-sensitive plants (Chapter 5, section 5.1). Therefore, it is essential to continue research at the cellular level for better understanding possible roles of ABA against LTI in roses. Further research is needed to investigate the most effective ABA concentrations, which will provide increased protection of roses against LTI without increasing senescence.

The effectiveness of ABA in inducing physiological responses that protect roses from LTI was tested using biochemical assays. Combined treatments with ABA before (pulse) and after storage (vase solution) prevented lipid peroxidation of rose petals. The presence of ABA either in pulsing or in vase solutions decreased electrolyte leakage both in petals and leaves of roses. ABA applied in vase solution was more effective than pulsed ABA treatment in reducing electrolyte leakage. Overall, these findings suggest that ABA treatment before or after low temperature storage might stabilise phospholipids or induce changes in membrane properties in roses as reported by Flores *et al.* (1988) for cucumber. However, additional research using a wide range of ABA concentrations may help to better understand the nature of ABA efficacy. Also, further study is required using biochemical assays to determine the process of membrane degradation (e.g. enzyme activity) in relation to ABA efficacy.

7.4 EFFECTS OF PBI-365 ON VASE LIFE OF ROSES STORED AT LOW TEMPERATURE

The use of wetting agents in roses has emerged in line with cut flower industry demand to prevent loss of quality during transport and extend vase life. For the first time, the synthetic ABA analogue PBI-365 was tested as a potential wetting agent for roses after storage. PBI-365 was effective in extending vase life after low temperature storage. Addition of 10^{-5} M PBI-365 in the vase solution decreased *ca.*

1.8- and 2.5-fold solution usage by roses with and without pulse ABA treatment before storage, respectively. Similarly, PBI-365 reduced vase solution usage by freshly harvested 'Baccara' roses during vase life (Pompodakis and Joyce, 2003). As synthetic ABA analogues are structurally similar compounds to the pure ABA (Abrams *et al.*, 1997), these positive effects of PBI-365 on vase life may be linked to the ability of ABA to induce stomatal closure. Also, PBI-365 was effective compared to untreated controls in reducing lipid peroxidation and, as a result, prevented electrolyte leakage after storage at 1°C. Thus, ABA analogues might have potential to *reduce LTI in roses. However, it is essential to continue investigations on a wider* range of PBI-365 concentrations that may be more effective in improving water relations before or after storage or during vase life evaluation. Additional research is required at the cellular level to explain the ABA analogue mode of action in roses.

7.5 FINAL CONCLUSION

The cut flower industry in the Mediterranean is economically dependent on rose exports to European countries during the year. This study was the first mention of vase life characteristics for roses in relation to seasonal changes in Mediterranean. This study has provided valuable insights into the environmental factors affecting postharvest quality of roses and potential to use ABA and its analogues in the cut rose industry. However, more research is needed to improve knowledge of the relationship between pre-harvest conditions in glasshouse and postharvest characteristics in roses. Investigation of ABA and analogue effects on vase life of different cut flower species remains an opportunity for future work.

REFERENCES

Abrams, S.R., Rose, P.A., Cutler, A.J., Balsevich, J.J., Lei, B. and Walker-Simmons, M.K. (1997) 8'-Methylene abscisic acid: An effective and persistent analog of abscisic acid. *Plant Physiology* **114**, 89-97.

Adachi, M., Kawabata, S. and Sakiyama, R. (2000) Effects of temperature and stem length on changes in carbohydrate content in summer-grown cut chrysanthemums during development and senescence. *Postharvest Biology and Technology* **20**, 63-70.

Anderson, M.D., Prasad, T.K., Martin, B.A. and Stewart, C.R. (1994) Differential gene expression in chilling acclimated maize seedlings and evidence for the involvement of abscisic acid in chilling tolerance. *Plant Physiology* **105**, 331-339.

Apelbaum, A. and Yang, S.F. (1981) Biosynthesis of stress ethylene induced by water deficit. *Plant Physiology* **68**, 594-596.

Aroca, R., Irigoyen, J.J. and Sanchez-Diaz, M. (2001) Photosynthetic characteristics and protective mechanisms against oxidative stress during chilling and subsequent recovery in two maize varieties differing in chilling sensitivity. *Plant science* **161**, 719-726.

Aroca, R., Vernieri, P., Irigoyen, Sanchez-Diaz, M., Tognoni, F. and Pardossi, A. (2003) Involvement of abscisic acid in leaf and root of maize (*Zea mays* L.) in avoiding chilling-induced water stress. *Plant Science* **165**, 671-679.

Awad, R.E., Meawad, A., Kamel Dawh, A. and El-Saha, M. (1986) Cut flower longevity as affected by chemical pre-treatment. *Acta Horticulturae* **181**, 177-182.

Baas, R. and Marissen, N. (2000) Cut rose quality as affected by calcium supply and translocation. *Acta Horticulturae* **518**, 45-54.

Baas, R., vanOers, S., Silber, A., Bernstein, N., Ioffe, M., Keinan, M. and Bar-Tal, A. (2003) Calcium distribution in cut roses as related to transpiration. *Journal of*

Horticultural Science and Biotechnology **78 (1)**, 1-9.

Barthe, P., Vaillant, V. and Gudin, S. (1991) Definition of indicators of senescence in the rose: Effect of the application of plant hormones. *Acta Horticulturae* **298**, 61-68.

Biolley, J.P. and Jay, M. (1993) Anthocyanins in modern roses: Chemical and colorimetric features in relation to the colour range. *Journal of Experimental Botany* **44**, 1725-1734.

Biran, I. and Halevy, A.H. (1974a) Effects of varying light intensities and temperature treatments applied to whole plants, or locally to leaves or flower buds, on growth pigmentation of 'Baccara' roses. *Physiologia Plantarum* **31**, 175-179.

Biran, I. and Halevy, A.H. (1974b) Effects of short-term heat and shade treatments on petal colour of 'Baccara' roses. *Physiologia Plantarum* **31**, 180-185.

Bleeksma, H.C. and van Doorn, W.G. (2003) Embolism in rose stems as a result of vascular occlusion by bacteria. *Postharvest Biology and Technology* 29, 334-340.

Boardman, N.K. (1977) Comparative photosynthesis of sun and shade plants. *Plant Physiology* **28**, 355-377.

Boese, S.R. Wolfe, D.W. and Melkonian, J.J. (1997) Elevated CO_2 mitigates chilling-induced water stress and photosynthetic reduction during chilling. *Plant, Cell and Environment* **20**, 625-632.

Borochov, A. and Woodson, W.R. (1989) Physiology and biochemistry of flower petal senescence. *Horticultural Review* **11**, 15-43.

Borochov, A., Mayak, S. and Halevy, A.H. (1976a) Combined effects of abscisic acid and sucrose on growth and senescence of rose flowers. *Plant Physiology* **36**, 221-224.

Borochov, A., Tirosh, T. and Halevy, A.H. (1976b) Abscisic acid content of

senescing petals on cut rose flowers as affected by sucrose and water stress. *Plant Physiology* **58**, 175-178.

Borochov, A., Mayak, S. and Broun, R. (1982) The involvement of water stress and ethylene in senescence of cut carnation flowers. *Journal of Experimental Botany* **33**, 1202-1209.

Bredmose, N. (1997) Year-round supplementary lighting at two photosynthetic photon flux densities for cut roses. *Acta Horticulturae* **418**, 59-64.

Brennan, R.M. and Jefferies, R.A. (1990) The use of chlorophyll fluorescence in assessment of low temperature hardiness in blackcurrant (*Ribes nigrum* L.). *Annals of Applied Biology* **117**, 667-672.

Burdett, A.N. (1970) The cause of bent neck in cut roses. *Journal of American Society for Horticultural Science* **95**, 427-431.

Cabrera, R.I. (2000) Evaluating yield and quality of roses with respect to nitrogen fertilization and leaf nitrogen status. *Acta Horticulturae* **511**, 133-140.

Cabrera, R.I. (2001) Effect of NaCl salinity and nitrogen fertilizer formulation on yield and nutrient status of roses. *Acta Horticulturae* **547**, 255-260.

Cabrera, R.I., Evans, R.Y. and Paul, J.L. (1993) Leaching losses of N from container-grown roses. *Scientia Horticulturae* **53**, 333-345.

Cameron, A.C. and Reid, M.S. (2001) 1-MCP blocks ethylene-induced petal abscission of *Pelargonium peltatum* but the effect is transient. *Postharvest Biology and Technology* **22**, 169-177.

Capell, B. and Dorffling, K. (1989) Low temperature-induced changes of abscisic acid contents in barley and cucumber leaves in relation to their water status. *Journal of Plant Physiology* **135**, 571-575.

Capell, B. and Dorffling, K. (1993) Genotype-specific differences of chilling tolerance of maize in relation to chilling-induced changes in water status and abscisic acid accumulation. *Physiologia Plantarum* **88**, 26-29.

Carpenter, W.J. and Rasmussen, H.P. (1974) The role of flower and leaves in cut flower water uptake. *Scientia Horticulturae* **2**, 293-298.

Carpenter, W.J. and Rasmussen, H.P. (1975) The role of flower and leaves in cut flower water uptake. *Floricultural Review* **156**, 24-25.

Carrasquer, A.M., Casals, I. and Alegre, L. (1990) Semi-automated method for the determination of abscisic acid in crude plant extracts. *Journal of Chromatography* **503**, 459-465.

Celikel, F.G. and Karacaly, Y. (1995) Effect of postharvest factors on flower quality and longevity of cut carnations (*Dianthus Caryophyllus* L.). *Acta Horticulturae* **405**, 156-163.

Chalker-Scott, L. (1999) Environmental significance of anthocyanins in plant stress responses. Invited review. *Photochemistry and Photobiology* **70 (1)**, 1-9.

Chandra, G., Reddy, K.S. and Mohan Ram, H.Y. (1981) Extension of vase life of cut marigold and chrysanthemum flowers by the use of cobalt chloride. *Indian Journal of Experimental Botany* **19**, 150-154

Chimonidou-Pavlidou, D. (1999) Irrigation and sensitive stages of rose development. *Acta Horticulturae* **481**, 393-401.

Chimonidou-Pavlidou, D. (2001) Effect of irrigation and shading at the stage of flower bud appearance. *Acta Horticulturae* **547**, 245-251.

Clark, G.E. and Burge, G.K. (1999) Effects of nitrogen nutrition on Sandersonia cut flower and tuber production in a soil-less medium. *New Zealand Journal of Crop and Horticultural Science* **27**, 145-152.

Cline, M.N. and Neely, D. (1983) The histology and histochemistry of the wound-healing process in *Geranium* cuttings. *HortScience* **108**, 779-780.

Come, D. (1991) Biological bases of the use of cold in ornamental horticulture. *Postharvest Physiology of Ornamentals* **298**, 21-28.

Cowan, I.R., Raven, J.A., Hartung, W. and Farquhar, G.D. (1982) A possible role for abscisic acid in coupling stomatal conductance and photosynthetic carbon metabolism in leaves. *Australian Journal of Plant Physiology* **9**, 489-498.

Dallaire, S., Houde, M., Gagne, Y., Saini, H.S., Boileau, S., Chevrier, N. and Sarhan, F. (1994) ABA and low temperature induce freezing tolerance via distinct regulatory pathways in wheat. *Plant Cell Physiology* **35**, 1-9.

Davies, W.J., Wilkinson, S. and Loveys, B. (2002) Stomatal control by chemical signalling and the exploitation of this mechanism to increase water use efficiency in agriculture. *New Phytologist* **153**, 449-460.

DeEll, J.R., Prange, R.K. and Murr, D.P. (1995) Chlorophyll fluorescence as a potential indicator of controlled-atmosphere disorders in 'Marshall' McIntosh apples. *HortScience* **30**, 1084-1085.

De La Guardia, M.D. and Benlloch, M. (1980) Effects of potassium and gibberellic acid on stem growth of whole sunflower plants. *Plant Physiology* **49**, 443-448.

Del Rio, M.A., Navarro, P. and Mateos, M. (1989) Effect of pre-treatment and storage conditions on cut rose flowers. *Acta Horticulturae* **246**, 319-325.

De Stigter, H.C.M. (1980) Water balance of cut and intact 'Sonia' rose plants. *Plant Physiology* **99**, 131-140.

De Witte, Y. and van Doorn, W.G. (1988) Identification of bacteria in the vase water of roses, and the effect of the isolated strains on water uptake. *Scientia Horticulturae*

35, 285-291.

De Witte, Y. and van Doorn, W.G. (1992) The mode of action of bacteria in the vascular occlusion of cut rose flowers. *Acta Horticulturae* **298**, 165-167.

Del Rio, M.A., Navarro, P. and Mateos, M. (1989) Effect of pretreatment and storage conditions on cut rose flowers. *Acta Horticulturae* **246**, 319-325.

Dieleman, J.A. (1996) Effect of cytokinins on axillary bud growth of 'Madelon' roses. *Acta Botany* **45**, 578-579.

Dilley, D.R. (1977) Hypobaric storage of perishable commodities-fruits, vegetables, flowers and seedlings. *Acta Horticulturae* **62**, 61-70.

Dixon, M.A. and Peterson, C.A. (1989) A re-examination of stem blockage in cut roses. *Scientia Horticulturae* **38**, 277-285.

Doi, M. and Reid, M.S. (1996) Postharvest characteristics of cut *Camellia japonica* L. 'Kumasaka'. *Postharvest Biology and Technology.* **7**, 331-340.

Donselman, H.M. and Broschat, T.K. Production of *Heliconia psittacorum* for cut flowers in South Florida. *Bulletin Heliconia Society Intstitute* **1**, 4-6.

Druege, U. (2001) Postharvest responses of different ornamental products to preharvest nitrogen supply: Role of carbohydrates, photosynthesis and plant hormones. *Acta Horticulturae* **543**, 97-105.

Druege, U., Zerche, S. and Kadner, R. (1998) Relation between nitrogen and soluble carbohydrate concentrations and subsequent rooting of chrysanthemum cuttings as influenced by nitrogen nutrition of stock plants and cool-storage of cuttings. *Acta Horticulturae* **517**, 81-89.

Drumm-Herrel, H. (1984) Blue/UV light effects on anthocyanin synthesis. In: Blue light effects in biological systems (Ed. Senger, H.). pp.375-383. Springer-Verlag,

Berlin.

Dufour, L. and Clairon, M. (1997) Advances in fertilization of *Anthurium* hybrid in Guadeloupe (F.W.I.). *Acta Horticulturae* **450**, 433-438.

Durkin, D.J. (1979a) Some characteristics of water flow through isolated rose stem segments. *Journal of American Society for Horticultural Science* **104**, 777-783.

Durkin, D.J. (1979b) Effect of millipore filtration, citric acid, and sucrose on peduncle water potential of cut rose flower. *HortScience* **104**, 860-863.

Eamus, D. (1987) Stomatal behaviour and leaf water potential of chilled and water-stressed *Solanum melongena*, as influenced by growth history. *Plant Cell Environment* **10**, 649-654.

Eamus, D. R., Fenton, R. and Wilson, J.M. (1983) Stomatal behaviour and water relations of chilled *Phaseolus vulgaris* L. and *Pisum sativum* L. *Journal of Experimental Botany* **34**, 434-441.

Elgar, H.J., Woolf, A.B. and Bieleski, R.L. (1999) Ethylene production of three lily species and their response to ethylene exposure. *Postharvest Biology and Technology* **16**, 257-267.

Even-Chen, Z. and Itai, C. (1975) The role of abscisic acid in senescence of detached tobacco leaves. *Physiologia Plantarum* **34**, 97-100.

Eze, J.M.O., Dumbroff, E.B. and Thompson, J.E. 1986) Effects of moisture stress and senescence on the synthesis of abscisic acid in the primary leaves of bean. *Physiologia Plantarum* **51**, 418-422.

Eze, J.M.O., Mayak, S., Thompson, J.E. and Dumbroff, E.B. (1986) Senescence in cut carnation flowers: Temporal and physiological relationships among water status, ethylene, abscisic acid and membrane permeability. *Physiologia Plantarum* **68**, 323-328.

Faragher, J.D. (1986) Postharvest physiology of waratah inflorescences (*Telopea speciosissima*, Proteaceae). *Scientia Horticulturae* **28**, 271-279.

Faragher, J.D. (1989) A review of research on postharvest physiology and horticulture of Australian native flowers. *Acta Horticulture* **261**, 249-256.

Faragher, J.D. and Mayak, S. (1984) Physiological responses of cut rose flowers to exposure to low temperature: Changes in membrane permeability and ethylene production. *Journal of Experimental Botany* **35 (156)**, 965-974.

Faragher, J.D., Mayak, S. and Tirosh, T. (1986) Physiological response of cut rose flowers to cold storage. *Plant Physiology* **67**, 205-210.

Faragher, J.D., Mayak, S., Tirosh, T. and Halevy, A.H. (1984) Cold storage of rose flowers: effects of cold storage and water loss on opening and vase life of 'Mercedes' roses. *Scientia Horticulturae* **24**, 369-378.

Feigin, A., Ginzburg, C., Gilead, S. and Ackerman, A. (1986) Effect of NH_4/NO_3 ratio in nutrient solution on growth and yield of greenhouse roses. *Acta Horticulturae* **189**, 127-135.

Field, A. (2000) Discovering statistics using SPSS for Windows. (Eds, Blakwell, G., de Leeuw, J., O' Muircheartaigh, C., Sarris, W., Schuman, H. and van Meter, K.), SAGE Publications, London, UK, pp.275.

Flexas, J., Briantais, J.M., Cerovic, Z., Medrano, H. and Moya, I. (2000) Steady-state and maximum chlorophyll fluorescence responses to water stress in grapevine leaves: A new remote sensing system. *Remote Sensing of Environment* **73**, 283-297.

Florack, D.E.A., Stiekema, W.J. and Bosch, D. (1996) Toxicity to peptides to bacteria present in the vase water of cut roses. *Postharvest Biology and Technology* **8**, 285-291.

Flores, A., Grau, A., Laurich, F. and Dorffling, K. (1998) Effect of new terpenoid analogues abscisic acid on chilling and freezing resistance. *Journal of Plant Physiology* **132**, 362-369.

Fujino, D.W., Reid, M.S. and Kohl, H.C. (1983) The water relations of Maidenhair fronds treated with silver nitrate. *Scientia Horticulturae* **19**, 349-355.

Garello, G., Menard, C., Dansereau, B. and Le Page-Degivry, M.T. (1995) The influence of light quality on rose flower senescence: involvement of abscisic acid. *Plant Growth Regulation* **16**, 135-139.

Gay, A.P. and Nichols, R. (1977) The effects of some chemical treatments on leaf water conductance of cut flowering stems of *Chrysanthemum morifolium*. *Scientia Horticulturae* **6**, 167-177.

Gislerod, H.R. (1999) The role of calcium on several aspects of plant and flower quality from a floricultural perspective. *Acta Horticulturae* **481**, 345-252.

Gislerod, H.R. and Mortensen, L.M. (1997) Effect of light intensity on growth and quality of cut roses. *Acta Horticulturae* **418**, 25-31.

Goszczynska, D.M. and Zieslin, N. (1993) Abscission of flower peduncles in rose (*Rosa* x *hybrida*) plants. *Scientia Horticulturae* **54**, 317-326.

Gowing, D.J.G, Jones, H.G. Davies, W.J. (1993) Xylem-transported abscisic acid: the relative importance of its mass and its concentration in the control of stomatal aperture. *Plant, Cell and Environment* **16**, 453-459.

Greenspan, L. (1977) Humidity fixed points of binary saturated aqueous solutions. *Journal of Research of the National Bureau of Standards. A. Physics and Chemistry* **81 (1)**, 89-96.

Hakam, N., Khanizadeh, S., DeEll, J.R. and Richer, C. (2000) Assessing chilling tolerance in roses using chlorophyll fluorescence. *HortScience* **35**, 184-186.

Halevy, A.H. (1972) Water stress and the timing of irrigation. *Horticultural Science* 7, 113-114.

Halevy, A.H. (1976) Treatments to improve water balance of cut flowers. *Acta Horticulturae* **64**, 223-230.

Halevy, A.H. and Mayak, S. (1975) Interrelationship of several phytohormones in the regulation of rose petal senescence. *Acta Horticulturae* **41**, 103-115.

Halevy, A.H. and Mayak, S. (1979) Senescence and postharvest physiology of cut flowers, Part 1. *Horticultural Reviews* **1**, 204-236.

Halevy, A.H. and Mayak, S. (1981) Senescence and postharvest physiology of cut flowers, Part 2. *Horticultural Reviews* **3**, 59-143.

Halevy, A.H., Byrne, T.G., Kofranek, A.M., Farnham, D.S. and Thompson, J.F. (1978) Evaluation of postharvest handling methods for transcontinental truck shipments of cut carnations, chrysanthemums, and roses. *Journal of American Society for Horticultural Science* **103**, 151-155.

Halevy, A.H., Mayak, S., Tirosh, T., Spiegelstein, H. and Kofranek, A.M. (1974) Opposing effects of abscisic acid on senescence of rose flowers. *Plant Physiology* **15**, 813-821.

Hammer, P.E. and Marois, J.J. (1989) Nonchemical methods for postharvest control of *Botrytis cinerea* on cut roses. *Phytopathology* **84**, 1305-1312.

Hanley, K.M. and Bramlage, W.J. (1989) Endogenous levels of abscisic acid in aging carnation flower parts. *Journal of Plant Growth Regulation* **8**, 225-236.

Hakam, N., Khanizadeh, S., Deell, J.R. and Richer, C. (2000) Assessing chilling tolerance in roses using chlorophyll fluorescence. *HortScience* **35**, 184-186.

Hartung, W., Wilkinson, S. and Davies, W.J. (1998) Factors that regulate abscisic acid concentrations at the primary site of action at the guard cell. *Journal of Experimental Botany* **49**, 361-367.

Hausladen, A. and Alscher, R.G. (1994) Cold hardiness-specific glutathione reductase isozymes in red spruce. *Plant Physiology* **105**, 215-223.

Heath, R.L. and Packer, L. (1968) Photoperoxidation in isolated chloroplasts. I. Kinetics and stoichiometry of fatty acid peroxidation. *Archives of Biochemistry and Biophysics* **125** (1), 189-198.

Hew, C.S., Lee, F.Y. and Wee, K.H. (1987) Factors affecting the longevity of Aranda flowers. *Acta Horticulturae* **205**, 195-202.

Ho, L.C. and Nichols, R. (1977) Translocation of ^{14}C-sucrose in relation to changes in carbohydrate content in rose corollas cut at different stages of development. *Annals of Botany* **41**, 227-242.

Hoogerwerf, A. and van Doorn, W.G. (1992) Numbers of bacteria in aqueous solutions used for postharvest handling of cut flowers. *Postharvest Biology and Technology* **1**, 295-304.

Hu, Y., Doi, M. and Imanishi, H. (1998a) Competitive water relations between leaves and flower bud during transport of cut roses. *Journal of Japanese Society for Horticultural Science* **67**, 532-536.

Hu, Y., Doi, M. and Imanishi, H. (1998b) Improving the longevity of cut roses by cool and wet transport. *Journal of Japanese Society for Horticultural Science* **67**, 681-684.

Hughes, H.E. and Hanan, J.J. (1978) Effect of salinity in water supplies on greenhouse rose production. *Journal of American Society for Horticultural Science* **103**, 694-699.

Ichimura, K., Shimamura, M. and Hisamatsu, T. (1998) Role of ethylene in senescence of cut *Eustoma* flowers. *Postharvest Biology and Technology* **14**, 193-198.

Janowiak, F. and Dorffling, K. (1996) Chilling of maize seedlings: changes in water status and abscisic acid content in ten genotypes differing in chilling tolerance. *Journal of Plant Physiology* **147**, 582-588.

Janowiak, F., Maas, B. and Dorffling, K. (2002) Importance of abscisic acid for chilling tolerance of maize seedlings. *Journal of Plant Physiology* **159**, 635-643.

Jeong-Seob, S., van Doorn, W.G. and Harkema, H. (1992) Water relations of cut rose flowers cv. Sonia after dry storage. *Journal of Korean Society for Horticultural Science* **33**, 337-342.

Jiao, J., Wang, X. and Tsujita, M.J. (1990) Whole plant net photosynthesis of miniature roses influenced by light, CO_2, and temperature. *Acta Horticulturae* **272**, 261-265.

Jones, R. and Faragher, J. (1991) Cold storage of selected members of the proteaceae and Australian native cut flowers. *HortScience* **26**, 1395-1397.

Jones, R. and Hill, M. (1993) The effect of germicides on the longevity of cut flowers. *Journal of American Society for Horticultural Science* **118**, 350-354.

Jones, R., Faragher, J.D. van Doorn, W.G. (1993) Water relations of cut flowering branches of Thryptomene calycina (Lindl.) Stapf. (Myrtaceae). *Postharvest Biology and Technology* **3**, 57-67.

Joyce, D.C. (1988) Postharvest characteristics of Geraldton waxflowers. *Journal of American Society for Horticultural Science* **13**, 748-742.

Joyce, D.C. (1989) Treatments to prevent flower abscission in Geraldton waxflower.

Hortscience **24**, 391.

Joyce, D.C. (1993) Postharvest floral organs fall in Geraldton waxflower (*Chamelaucium uncinatum* Schauer). *Australian Journal of Experimental Agriculture* **33**, 481-487.

Joyce, D.C. and Jones, P.N. (1992) Water balance of the foliage of cut Geraldton waxflower. *Postharvest Biology and Technology* **2**, 31-39.

Joyce, D.C. and Shorter, A.J. (2000) Long term, low temperature storage injures kangaroo paw cut flowers. *Postharvest Biology and Technology* **20**, 203-206.

Joyce, D.C., Meara, S.A., Hetherington, S.E. and Jones, P.N. (2000) Effects of cold storage on cut *Grevillea* 'Sylvia' inflorescences. *Postharvest Biology and Technology* **18**, 49-56.

Joyce, D.C., Shorter, A.J. and Jones, P.N. (1996) A triazole compound extends the vase life of Geraldton waxflower. *Australian Journal of Experimental Agriculture* **36**, 117-119.

Joyce, D., Jones, R. and Faragher, J. (1993) Postharvest characteristics of native Australian flowers. *Postharvest News and Information* **4**, 61-67.

Karabal, E., Yucel, M. and Oktem H.A. (2003) Antioxidant responses of tolerant and sensitive barley cultivars to boron toxicity. *Plant Science* **164 (6)**, 925-933.

Ketsa, S. and Boonrote, A. (1990) Holding solutions for maximizing bud opening and vase-life of Dendrobium 'Youppadeewan' flowers. *Journal of Horticultural Science* **65**, 41-47.

King, A.I., Joyce, D.C. and Reid, M.S. (1988) Role of carbohydrates in diurnal chilling sensitivity of tomato seedlings. *Plant Physiology* **86**, 764-768.

Knee, M. (1995) Copper reverses silver inhibition of flower senescence in *Petunia*

hybrida. Postharvest Biology and Technology **6**, 121-128.

Knee, M. (2000) Selection of biocides for use in floral preservatives. *Postharvest Biology and Technology* **18**, 227-234.

Kofranek, A.M., Kohl. H. C. and Kubota, J. (1974) A slow-release chlorine compound as a vase water additive. *Floricultural Review* **154**, 63-65.

Kofranek, A.M., Evans, E., Kubota, J. and Farnham, D.S. (1978) Chemical pre-treatments for China asters to increase flower longevity. *Florists' Review* **162**, 70-72.

Kohl, H.C. (1961) Rose neck droop. *California State Floricultural Association.* **10**, 4-5.

Kohl, H.C. and Rundle, D.L. (1972) Decreasing water loss of cut roses with abscisic acid. *HortScience* **7**, 249.

Kramer, P.J. (1981) Carbon dioxide concentration, photosynthesis, and dry matter production. *Biological Science* **31**, 29-33.

Krause, G.H. and Weis, E. (1984) Chlorophyll fluorescence as a tool in plant physiology. II. Interpretation of fluorescence signals. *Photosynthesis Research* **5**, 139-157.

Krause, G.H. and Weis, E. (1991) Chlorophyll fluorescence and photosynthesis: The basics. *Annual Review of Plant Physiology and Molecular Biology* **42,** 313-349.

Kuiper, D., Van Reenen, H. S. and Ribot, S. A. (1996) Characterisation of flower bud opening in roses; a comparison of 'Madelon' and 'Sonia' roses. *Postharvest Biology and Technology* **9**, 75-86.

Lamb, N., Wahab, N., Rose, P.A., Shaw, A.C., Abrams, S.R., Cutler, A.J., Smith, P.J., Gusta, L.V. and Ewan, B. (1996) Synthesis metabolism and biological activity of a

deuterated analogue of the plant hormone S-(+)-abscisic acid. *Phytochemistry* **41**, 23-28.

Lee, T.M., Lur, H.S. and Chu, C. (1997) Role of abscisic acid in chilling tolerance of rice (*Oryza sativa* L.). II Modulation of free polyamine levels. *Plant Science* **126**, 1-10.

Leonard, R.T., Nell, T.A., Suzuki, A., Barrett, J.E. and Clark, D.G. (2001) Evaluation of long term transport of Colombian growth cut roses. *Acta Horticulturae* **543**, 293-297.

Le Page-Degivry, M.T., Orlandini, M., Garello, G., Barthe, P. and Gudin, S. (1991) Regulation of ABA levels is senescing petals of rose flowers. *Journal of Plant Growth Regulation* **10**, 67-72.

Li, P.H. (1994) Maize chilling tolerance induction. In: Crop adaptation to cool climates. p. 579-594. European Commission, Brussels.

Li, P.H., Chen, W.P., Jian, L. and Xin, Z. (1997) Abscisic acid-induced chilling tolerance in maize. In: Plant cold hardiness. Molecular Biology, biochemistry and physiology. p. 215-223. Plenum Press, New York and London.

Lineberger, R.D. and Steponkus, P.L. (1976) Identification and localization of vascular occlusions in cut roses. *Journal of American Society for Horticultural Science* **101**, 246-250.

Lipari, V. and Romano, D. (1989) Production results of the carnation cultivated in a cold greenhouse. *Acta Horticulturae* **246**, 139-143.

Little, T.M. (1985) Analysis of percentage and rating scale data. *HortScience* **20**, 642-644.

Ludewig, M., Dorffling, H. Siefert, H. (1988) Abscisic acid and water transport in sunflowers. *Planta* **175**, 325-333.

Lurie, S., Ronen, R. and Meier, S. (1994) Determining chilling injury induction in green peppers using nondestructive pulse amplitude modulated (PAM) fluorometry. *Journal of American Society for Horticultural Science* **119**, 59-62.

Maas, F.M. and Bakx, E.J. (1997) Growth and flower development in roses as affected by light. *Acta Horticulturae* **418**, 127-134.

Macnish, A.J., Joyce, D.C., Hofman, P.J., Simons, D.H. and Reid, M.S. (2000a) 1-Methylcyclopropene treatment efficacy in preventing ethylene perception in banana fruit and grevillea and waxflower flowers. *Australian Journal of Experimental Agriculture* **40**, 471-481.

Macnish, A.J., Simons, D.H., Joyce, D.C., Faragher, J.D. and Hofman, P.J. (2000b) Responses of native Australian cut flowers to treatment with 1-Methylcyclopropene and ethylene. *HortScience* **35 (2)**, 254-255.

Malshet, V.G. and Tappel, A.L. (1973) Fluorescent products of lipid peroxidation: I. Structural requirement for fluorescence in conjugated Schiff's bases. *Lipids* **8**, 194-198.

Marangoni, A.G., Palma, T. and Stanley, D.W. (1996) Membrane effects in postharvest physiology. *Postharvest Biology and Technology* **7**, 193-217.

Marissen, N. (2001) Effects of pre-harvest light intensity and temperature on carbohydrate levels and vase life of cut roses. *Acta Horticulturae* **543**, 331-336.

Markhart, A.H. (1984) Amelioration of chilling-induced water stress by abscisic acid-induced changes in root hydraulic conductance. *Plant Physiology* **74**, 81-83.

Markhart, A.H. and Harper, M.S. (1995) Deleterious effects of sucrose in preservatives solutions on leaves of cut roses. *HortScience* **30**, 1429-1432.

Marousky, F.J. (1969) Vascular blockage, water absorption stomatal opening and

respiration of cut 'Better Times' roses treated with 8-hydroxiquinoline citrate and sucrose. *Journal of American Society for Horticultural Science* **94**, 223-226.

Marousky, F.J. (1971) Inhibition of vascular blockage and increased moisture retention in cut roses induced by pH, 8-Hydroxyquinoline citrate, and sucrose. *Journal of American Society for Horticultural Science* **96**, 38-41.

Marousky, F.J. (1976) Control of bacteria in vase water and quality of cut flowers as influenced by sodium dichloroisocyanurate, 1,3-dichloro-5,5-dimethylhydantoin and sucrose. *USDA, Agricultural Research Service* S-115.

Maxwell, K. and Johnson, G.N. (2000) Chlorophyll fluorescence – a practical guide. *Journal of Experimental Botany* **51**, 659-668.

Mayak, S. and Dilley, D.R. (1976) Effect of sucrose on response of cut carnation to kinetin, ethylene and abscisic acid. *HortScience* **101**, 583-585.

Mayak, S. and Faragher, J.D. (1986) Storage environment related stresses and flower senescence. *Acta Horticulturae* **181**, 33-43.

Mayak, S. and Halevy, A.H. (1972) Interrelationships of ethylene and abscisic acid in the control of rose petal senescence. *Plant Physiology* **50**, 341-346.

Mayak, S. and Halevy, A.H. (1974) The action of kinetin in improving the water balance and delaying senescence processes of cut rose flowers. *Physiologia Plantarum* **32**, 330-336.

Mayak, S., Meir, S. and Ben-Sade, H. (2001) The effect of transient water stress on sugar metabolism and development of cut flowers. *Acta Horticulturae* **543**, 191-197.

Mayak, S., Borochov, A. and Tirosh, T. (1985) Transient water stress in carnation flowers: Effect of amino-oxyacetic acid. *Journal of Experimental Botany* **36**, 800-806.

Mayak, S., Halevy, A.H., Sagie, S., Bar-Yoseph, A. and Bravdo, B. (1974) The water balance of cut rose flowers. *Plant Physiology* **31**, 15-22.

McGuire, R.G. (1992). Reporting of objective colour measurements. *HortScience* **27**, 1254-1255.

Meir, S., Rubin, L., Zauberman, G. and Fuchs, Y. (1992) Changes in fluorescence lipid peroxidation products of room-ripened and vine-ripened tomato fruits in relation to other ripening parameters. *Postharvest Biology and Technology* **2**, 125-235.

Menard, C., Dansereau, B., Garello, G. and Le Page-Degivry, M.T. (1995) Influence of nitrogen supply on ABA levels and flower senescence in *Rosa hybrida* cv. Royalty. *Acta Horticulturae* **424**, 151-153.

Miranda, J.H., Joyce, D.C., Hetherington, S.E. and Jones, P.N. (2000) Cold-storage-induced changes in chlorophyll fluorescence of kangaroo paw Bush Dawn flowers. *Australian Journal of Experimental Agriculture* **40**, 1151-1155.

Moe, R. (1975) The effect of growing temperature on keeping quality of cut roses. *Acta Horticulturae* **41**, 77-93.

Moe, R. (1988) Growth and flowering in roses. *Acta Horticulturae* **218**, 121-130.

Moe, R. and Kristoffersen, T. (1969) The effect of temperature and light on growth and flowering of Rosa 'Baccara' in greenhouses. *Acta Horticulturae* **14**, 157-167.

Mohan Ram, H.Y. and Rao, I.V. (1977) Prolongation of vase life of *Lupinus hartwegii* Lindl. by chemical treatments. *Scientia Horticulturae* **7**, 377-382.

Montero, E., Sibole, J., Cabot, C., Poschenrieder, C. and Barcelo, J. (1994) Abscisic acid content of salt-stressed *Phaseolus vulgaris* L. – Comparison of high-performance-liquid-chromatography, gas-chromatography with electron-capture detection, enzyme-linked-immunosorbent-assay and radioimmunoassay. *Journal of Chromatography A* **658 (1),** 83-90.

Mortensen, L.M. (2001) Greenhouse climate and keeping quality of roses. *Acta Horticulturae* **543**, 199-205.

Mortensen, L.M. and Fjeld, T. (1995) High air humidity reduces the keeping quality of cut roses. *Acta Horticulturae* **405**, 148-155.

Mortensen, L.M. and Fjeld, T. (1998) Effects of air humidity, lighting period and lamp type on growth and vase life of roses. *Scientia Horticulturae* **73**, 229-237.

Mortensen, L.M. and Gislerod, H.R. (1999) Influence of air humidity and lighting period on growth, vase life and water relations of 14 rose cultivars. *Scientia Horticulturae* **82**, 289-298.

Mortensen, L.M., Gislerod, H.R. and Mikkelsen, H. (1992) Maximizing the yield of greenhouse roses with respect to artificial lighting. *Norwegian Journal of Agricultural Sciences* **6**, 27-34.

Mortensen, L.M., Ottosen, C. and Gislerod, H.R. (2001) Effects of air humidity and K:Ca ratio on growth, morphology, flowering and keeping quality of pot roses. *Scientia Horticulturae* **90**, 131-141.

Muller, R., Andersen, A.S. and Serek, M. (1998) Differences in display life of miniature potted roses (*Rosa hybrida* L.). *Scientia Horticulturae* **76**, 59-71.

Muller, R., Stummann, B.M., Andersen, A.S. and Serek, M. (1999) Involvement of ABA in postharvest life of miniature potted roses. *Plant Growth Regulation* **29**, 143-150.

Muller, R., Lind-Iversen, S., Stummann, B.M. and Serek, M. (2000) Expression of genes for ethylene biosynthetic enzymes and an ethylene receptor in senescing flowers of miniature potted roses. *Journal of Horticultural Science and Biotechnology* **75**, 12-18.

Muller, R., Stummann, B.M. and Andersen, A.S. (2001) Comparison of postharvest properties of closely related miniature rose cultivars (*Rosa hybrida* L.). *Scientia Horticulturae* **91**, 325-338.

Murata, T. (1989) Relation of chilling stress to membrane permeability. p. 201-209. In: Chilling injury of horticultural crops. CRC Press, Boca Raton.

Murr, D.P., Venkatarayappa, T. and Tsujita, M.J. (1979) Counteraction of bent-neck of cut roses with cobalt nitrate. *Canadian Journal of Plant Science* **59**, 1169-1171.

Nichols, R. (1973) Senescence of cut carnation flower: respiration and sugar status. *HortScience* **48**, 111-121.

Nichols, R. (1975) Chrysanthemum (*Chrysanthemum morifolium* Ramat.): solution uptake and flower quality. p. 50-51. In: Annual Report 1974, Glasshouse Crop Research Institute of Littlehampton, UK.

Nowak, J. (2001) The effect of phosphorus nutrition on growth, flowering and leaf nutrient concentrations of osteospermum. *Acta Horticulturae* **548**, 557-560.

Nowak, J. and Rudnicki, R.M. (1990) Postharvest handling and storage of cut flowers, florist greens, and potted plants. Timber Press, Inc., Portland.

Ohkawa, K., Kasahara, Y. and Suh, J.N. (1999) Mobility and effects on vase life of silver-containing compounds in cut rose flowers. *HortScience* **34**, 112-113.

Oren-Shamir, M., Dela, G., Ovadia, R., Nissim-Levi, A., Philosoph-Hadas, S. and Meir, S. (2001). Differentiation between petal blueing and senescence of cut 'Mercedes' rose flowers. *Journal of Horticultural Science and Biotechnology* **76 (2)**, 195-200.

Orlandini, M., Arene, L. and Le Page-Degivry, M.T. (1991) The relationship between petal water potential and levels of abscisic acid in rose flower. *Acta Horticulturae* **298**, 161-163.

Panavas, T. and Rubinstein, B. (1998) Oxidative events during programmed cell death of daylily (*Hemerocallis* hybrid) petals. *Plant Science* **133,** 125-138.

Papadimitriou, M. (1995) Pre- and postharvest treatments on vase life of 'Sonia' and 'Madelon' roses. PhD Thesis, Department of Agricultural Science, Aristotile univeristy of Thessaloniki, Greece.

Panavas, T. and Rubinstein, B. (1998) Oxidative events during programmed cell death of daylily (*Hemerocallis* hybrid) petals. *Plant Science* **133,** 125-138.

Pardossi, A., Vernieri, P. and Tognoni, F. (1992) Involvement of abscisic acid in regulating water status in *Phaseolus vulgaris* L. during chilling. *Plant Physiology* **100**, 1243-1250.

Parups, E.V. and Voisey, P.W. (1976) Lignin content and resistance ot bending of the pedicel in greenhouse-grown roses. *Journal of Horticultural Science* **51**, 253-259.

Paulin, A. (1975) Effects of watering following a drought period on nitrogen metabolism of cut *Iris germanica* flowers. *Acta Horticulturae* **41**, 13-20.

Pearce, R.S. (1999) Molecular analysis of acclimation to cold. *Plant Growth Regulation* **29**, 47-126.

Philosoph-Hadas, S., Hadas, E. and Aharoni, N. (1993) Characterization and use in ELISA of a new monoclonal antibody for quantitation of abscisic acid in senescing rice leaves. *Plant Growth Regulation* **12**, 71-78.

Piqueras, A., Cortina, M., Serna, M.D. and Casas, J.L. (2002) Polyamines and hyperhydricity in micropropagated carnation plants. *Plant Science* **162,** 671-678.

Platt-Aloia, K.A. and Thomson, W.W. (1987) Freeze fracture evidence for lateral phase separations in the plasmalemma of chilling-injured avocado fruit. *Protoplasma* **136**, 71-80.

Pompodakis, N. E. and Joyce, D. C. (2003) ABA analogue effects on vase life and leaf crisping of cut 'Baccara' roses. *Australian Journal of Experimental Agriculture* **43**, 425-428.

Pompodakis, N.E., Joyce, D.C., Terry, L.A. and Lydakis, D.E. (2004) Effects of vase solution pH and abscisic acid on the longevity of cut 'Baccara' roses. *Journal of Horticultural Science and Biotechnology* **79**, 828-832.

Pompodakis, N.E., Terry, L.A., Joyce, D.C., Lydakis, D.E. and Papadimitriou, M.E. (2005) Effects of seasonal variation and low temperature storage on vase life and leaf chlorophyll fluorescence of roses. *Postharvest Biology and Technology*, in press.

Porat, R., Shlomo, E., Serek, M., Sisler, E.C. and Borochov, A. (1995) 1-Methylcyclopropene inhibits ethylene action in cut phlox flowers. *Postharvest Biology and Technology* **6**, 313-319.

Prasad, T.K., Anderson, M.D. and Stewart, C.R. (1994) Acclimation, hydrogen peroxide, and abscisic acid protect mitochondria against irreversible chilling injury in maize seedlings. *Plant Physiology* **105**, 619-627.

Purvis, M.J., Collier, D.C. and Walls, D. (1966) Laboratory techniques in botany, second edition, London, Butterworths.

Put, H.M.C. and Clerkx, A.C.M. (1988) The infiltration ability of micro-organisms *Bacillus*, *Fusarium*, *Kluyveromyces* and *Pseudomonas* spp. into xylem vessels of Gerbera cv. 'Fleur' and Rosa cv. 'Sonia' cut flowers: a scanning electron microscope study. *Journal of Applied Bacteriology* **64**, 515-530.

Put, H.M.C. and Jansen, L. (1989) The effects on the vase life of cut *Rosa* cultivar 'Sonia' of bacteria added to the vase water . *Scientia Horticulturae* **39**, 167-179.

Put, H.M.C. and Klop, W. (1990) The effects of microbial exopolysaccharides (EPS) in vase water on the water relations and the vase life of *Rosa* cv. Sonia. *Journal of*

Applied Bacteriology **68**, 367-384.

Put, H.M.C. and Rombouts, F.M. (1989) The influence of purified microbial pectic enzymes on the xylem anatomy, water uptake and vase life of *Rosa* cultivar 'Sonia'. *Scientia Horticulturae* **38**, 147-160.

Raison, J.K. and Lyons, J.M. (1986) Chilling injury: A plea for uniform terminology. *Plant, Cell and Environment* **9**, 685-686.

Raison, J.K. and Orr, G.R. (1986) Phase transitions in liposomes formed from the polar lipids of mitochondria from chilling sensitive plants. *Plant Physiology* **81**, 807-811.

Rajapakse, N.C., Kelly, J.W. and Miller, W.B. (1997) Influence of carbon dioxide enrichment on postharvest leaf chlorosis of miniature roses. *Department of Horticulturae, Clemson University, USA.*

Reddy, T.V. (1988) Mode of action of cobalt extending the vase life of cut roses. *Scientia Horticulturae* **36**, 303-313.

Reddy, T.V., Nagarajaiah, C. and Raju, B. (1988) Impregnating cut rose stems with nickel increases vase life. *Current Research, University of Agricultural Science, Bangalore* **17**, 108-109.

Redman, P.B. Dole, J.M., Maness, N.O. and Anderson, J.A. (2002) Postharvest handling of nine specialty cut flower species. *Scientia Horticulturae* **92**, 293-303.

Reid, M.S., Evans, R.Y., Dodge, L.L. and Mor, Y. (1989a) Ethylene and silver thiosulfate influence opening of cut rose flowers. *HortScience* **114**, 436-440.

Reid, M.S., Evans, R.Y., Dodge, L.L. and Mor, Y. (1989b) Effects of ethylene on rose opening. *Acta Horticulturae* **261**, 215-220.

Ristic, Z., Yang, G., Sterzinger, A. and Zhang L. (1998) Higher chilling tolerance in maize is not always related to the ability for greater and faster abscisic acid accumulation. *Journal of Plant Physiology* **153**, 154-162.

Rogers, M.N. (1973) An historical and critical review of postharvest physiology research on cut flowers. *HortScience* **8**, 189-194.

Rose, O.A., Cutler, A.J., Irvine, N.M., Shaw, A.C., Squires, T.M., Loewen, M.K. and Abrams, S.R. (1997) 8'-acetylene ABA: An irreversible inhibitor of ABA 8'-hydroxylase. *Bioorganic and Medical Chemistry Letters* 7, 2543-2545.

Rosenqvist, E. and van Kooten, O. (2003) Chlorophyll fluorescence: A general description and nomenclature, p. 31-77. In: DeEll, J.R and Toivonen, P.M.A. (Editors). Practical applications of chlorophyll fluorescence in plant biology. Kluwer Academic Publishers, New York.

Roychowdhury, N. and Roychowdhury, P. (1995) The effect of field application of K on postharvest behaviour of gladiolus. *Acta Horticulturae* **405**, 170-172.

Ruamrungsri, S., Ikarashi, T. and Ohyama, T. (1997) Uptake, translocation and fractionation of nitrogen in narcissus organs by using ^{15}N. *Acta Horticulturae* **430**, 73-78.

Rubinstein, B. (2000) Regulation of cell death in flower petals. *Plant Molecular Biology* **44**, 303-318.

Ruting, A. (1991) Effects of wetting agents and cut flower food on the vase life of cut roses. *Acta Horticulturae* **298**, 69-73.

Ruzin, S.E. (1999) Plant Microtechnique and Microscopy. Oxford University Press, pp. 93-95.

Ryan, W.L. (1957) Flower preservatives: using silver and zinc ions as disinfectants. *Florists' Review* **121**, 59-60.

Salisbury, F.B. and Ross, C.W (1992) Plant Physiology (Book), Fourth Edition. Wadsworth Publishing Co., Belmont, CA.

Salter, P.J. and Goode, J.E. (1967) Crop responses to water at different stages of growth. *UK: Commonwealth Agricultural Bureau* pp., 205.

Sancho, A.J.F. (1989) Environmental influences on growth of ornamental plants. *Acta Horticulturae* **246**, 79-96.

Sarkka, L.E., Rita, H.J. and Ripatti, S.O. (2001) Cut rose flower longevity and its variation between plants of cv. Frisco grown in different lighting periods. *Acta Horticulturae* **547**, 261-268.

Schapendonk, A.H.C.M., Van Der Putter, P.E.L., Dolstra, O., Haalstra, S.R. and Tonk, W.J.M. (1992) Chlorophyll fluorescence: a non-destructive method for detecting damage in the photosynthesis apparatus in plants. *Acta Horticulturae* **304**, 61-70.

Scholes, J. and Boodley, J.W. (1964) Improved lasting life of 'Velvet Times' roses with chemicals. *New York State Flower Growers Bulletin* **224**, 1-4.

Sen Gupta, A., Webb, R.P., Holaday, A.S. and Allen, R.D. (1993) Overexpression of superoxide dismutase protects plants from oxidative stress. Induction of ascorbate peroxidase in superoxide dismutase-overexpressing plants. *Plant Physiology* **103**, 1067-1073.

Serek, M. (1990) Effects of pre-harvest fertilization on the flower longevity of potted *Campanula carpatica* 'Karl Foerster'. *Scientia Horticulturae* **44**, 119-126.

Serek, M. and Sisler, E.C. (2001) Efficacy of inhibitors of ethylene binding in improvement of the postharvest characteristics of potted flowering plants. *Postharvest Biology and Technology* **23**, 161-166.

Serek, M., Sisler, E.C. and Reid, M.S. (1994) Novel gaseous ethylene binding inhibitor prevents ethylene effects in potted flowering plants. *Journal of American Society for Horticultural Science* **119**, 1230-1233.

Serrano, M., Amoros, A., Pretel, M.T., Martinez-Madrid, M.C. and Romojaro, F. (2001) Preservative solutions containing boric acid delay senescence of carnation flowers. *Postharvest Biology and Technology* **23**, 133-142.

Serrano, M., Martinez, G., Pretel, M.T., Riquelme, F. and Romojaro, F. (1992) Cold storage of rose flowers (*Rosa hybrida*, cultivar 'Visa'): physiological alternations. *Scientia Horticulturae* **51**, 129-137.

Serrano, M., Martinez-Madrid, M.C., Riquelme, F. and Romojaro, F. (1995) Enhanced ethylene synthesis in cold stored carnation flowers. *Acta Horticulturae* **405**, 298- 305.

Sevelius, N., Hyttinen, T. and Somersalo, S. (2001) Selection of rose cultivars for low light greenhouse production by photosynthetic features. *Acta Horticulturae* **547**, 159-166.

Sexton, R. and Porter, A.E., Littlejohns, S. and Thain, S.C. Effects of Diazocyclopentadiene (DACP) and silver thiosulphate (STS) on ethylene regulated abscission of sweet pea flowers (*Lathyrus odoratus* L.). *Annals of Botany* **75**, 337-342.

Sharom, M., Willemot, C. and Thompson, J.E. (1994) Chilling injury induces lipis phase changes in membranes of tomato fruit. *Plant Physiology* **105**, 305-308.

Shin, H.K., Lieth, J.H. and Kim, S.H. (2001) Effects of temperature on leaf area and flower size in rose. *Acta Horticulturae* **547**, 185-191.

Singh, K. and Moore, K.G. (1992) Water relations of cut chrysanthemum flowers. *HortScience* **6**, 121-124.

Slootweg, G. and Van Meeteren, U. (1991) Transpiration and stomatal conductance of roses cv. Sonia grown with supplemental lighting. *Acta Horticulturae* **298**, 119-125.

Slootweg, G., Ten Hoope, M. and De Gelder, A. (2001) Seasonal changes in vase life, transpiration and leaf drying of cut roses. *Acta Horticulturae* **543**, 337-342.

Smillie, R.M. and Hetherington, S.E. (1990) Screening for stress tolerance by chlorophyll fluorescence, p. 229-261. In: Hashimoto, H., Nonami, H., Kramer, P.J. and Strain, B.R. (Editors). Measurement techniques in plant science. Academic, San Diego.

Sood, S. and Nagar, P.K. (2003) Changes in abscisic acid and phenols during flower development in two diverse species of rose. *Acta Physiologiae Plantarum* **25 (4)**, 411-416.

Spikman, G. (1986) The effect of water stress on ethylene production and ethylene sensitivity of freesia inflorescences. *Acta Horticulturae* **181**, 135-140.

Spychalla, J.P. and Desborough, S.L. (1990) Superoxide dismutase, catalase, α-tocopherol content of stored potato tubers. *Plant Physiology* **94**, 1214-1218.

Staby, G.L., Cunningham, M.S., Holstead, C.L., Kelly, J.W., Konjoian, P.S., Eisenberg, B.A. and Dressler, B.S. (1984) *Journal of American Society for Horticultural Science* **109 (2)**, 193-197.

Starck, Z., Choluj, D. and Gawronska, H. (1998) The effect of drought hardening and chilling on ABA content in xylem sap and ABA delivery rate from root of tomato plant. *Acta Physiology Plantarum* **20**, 41-48.

Stubbs, J.M. and Francis, M.J.O. (1971) Electron microscopical studies of rose petal cells during flower maturation. *Planta Medica* **20**, 211-218.

Sung, J.M. and Jeng, T.L. (1994) Lipid peroxidation and peroxide-scavenging

enzymes associated with accelerated aging pf peanut seed. *Physiologia Plantarum* **91**, 51-55.

Sytsema-Kalkman, E.C. (1991) Postharvest studies on *Syringa vulgaris*. *Acta Horticulturae* **298**, 127-133.

Taiz, L. and Zeiger, E. (1998) Plant Physiology (Book). Sinauer Associates, Inc., Publishers, Sunderland, MA.

Tamminen, I., Makela, P., Heino, P. and Palva, E.T. (2001) Ectopic expression of *ABI3* gene enhances freezing tolerance in response to abscisic acid and low temperature in *Arabidopsis thaliana*. *Plant Journal* **25**, 1-8.

Tantau, H. Dorffling, K. (1991) Effects of chilling on physiological responses and changes in hormone levels in two *Euphorbia pulcherrima* varieties with different chilling tolerance. *Journal of Plant Physiology* **138**, 734-740.

Ter Hell, B. and Hendriks, L. (1995) The influence of nitrogen on keeping quality of pot plants. *Acta Horticulturae* **405**, 138-141.

Terry, L.A., Joyce, D.C. and Khambay, B.P.S. (2003) Antifungal compounds in Geraldton waxflower tissues. *Australasian Plant Pathology* **32,** 411-420.

Tjia, B.O. and Funnel, K.A. (1986) Postharvest studies of cut Zantedeschia inflorescences. *Acta Horticulturae* **181**, 451-457.

Torre, S. and Fjeld, T. (2001) Water loss and post-harvest characteristics of cut roses grown at high or moderate relative air humidity. *Scientia Horticulturae* **89**, 217-226.

Torre, S., Fjeld, T. and Gislerod, H.R. (2001) Effects of air humidity and K/Ca ratio in the nutrient supply on growth and postharvest characteristics of cut roses. *Scientia Horticulturae* **90**, 291-304.

Trejo, C.L., Davies, W.J. and Ruiz, L.M.P. (1993) Sensitivity of stomata to abscisic

acid. *Plant Physiology* **102**, 497- 502.

Trouverie, J., Thevenot, C., Rocher, J.P., Sotta, B. and Prioul, J.L. (2003) The role of abscisic acid in the response of a specific vacuolar invertase to water stress in the adult maize leaf. *Journal of Experimental Botany* **54**, 2177-2186.

Ueda, Y., Nishihara, S., Tomita, H. and Oda, Y. (2000) Photosynthetic response of Japanese rose species *Rosa bracteata* and *Rosa rugosa* to temperature and light. *Scientia Horticulturae* **84**, 365-371.

Urban, L., Fabret, C. and Barthelemy, L. (1998) Changes in stem diameter depend upon variations in water content in rose plants. *Acta Horticulturae* **424**, 67-72.

van Doorn, W.G. (1995) Vascular occlusion in cut rose flowers: A survey. *Acta Horticulturae* **405**, 58-66.

van Doorn, W.G. (1997) Water relations of cut flowers. *Horticultural Reviews* **18**, 1-85.

van Doorn, W.G. (2001) Categories of petal senescence and abscission: A re-evaluation. *Annals of Botany* **87**, 447-456.

van Doorn, W.G. and Cruz, P. (2000) Evidence for a wounding-induced xylem occlusion in stems of cut chrysanthemum flowers. *Postharvest Biology and Technology* **19**, 73-83.

van Doorn, W.G. and De Witte, Y. (1991a) Effect of dry storage on bacterial counts in stems of cut rose flowers. *Horticultural Science* **26**, 1521-1522.

van Doorn, W.G. and De Witte, Y. (1991b) Effect of bacterial suspensions on vascular occlusion in stems of cut rose flowers. *Journal of Applied Bacteriology* **71**, 119-123.

van Doorn, W.G. and Jones, R.B. (1994) Ultrasonic acoustic emissions from excised

stems of two *Thryptomene* species. *Physiologia Plantarum* **92**, 431-436.

van Doorn, W.G. and Otma, E. (1994) Vascular occlusion in cut flowering rose stems exposed to air: Role of water entry into the lumina of the xylem conduits opened by cutting. *Plant Physiology* **145**, 78-82.

van Doorn, W.G. and Perik, R.J.J. (1990) Hydroxyquinoline citrate and low pH prevent vascular blockage in stems of cut rose flowers by reducing the number of bacteria. *Journal of American Society for Horticultural Science* **115**, 979-981.

van Doorn, W.G. and Reid, M.S. (1995) Vascular occlusion in stems of cut rose flowers exposed to air: Role of xylem anatomy and rates of transpiration. *Physiologia Plantarum* **93**, 624-629.

van Doorn, W.G. and Suiro, V. (1996) Relationship between cavitation and water uptake in rose stems. *Physiologia Plantarum* **96**, 305-311.

van Doorn, W.G. and Vojinovic, A. (1996) Petal abscission in rose flowers: Effects of water potential, light intensity and light quality. *Annals of Botany* **78**, 619-623.

van Doorn, W.G., Bakker, L. and Veken, M. (1994) Effect of dry storage on scape bending in cut *Gerbera jamesonii* flowers. *Postharvest Biology and Technology* **4**, 261-269.

van Doorn, W.G., Clerkx, A.C.M. and Boekestein, A. (1991a) Bacteria as a cause of vascular occlusion in cut fronds of *Adiantum raddianum*: a scanning electron microscope study. *Scientia Horticulturae* **48**, 299-309.

van Doorn, W.G., Groenewegen, G. and van de Pol, P.A. (1991b) Effects of carbohydrate and water status on flower opening of cut 'Madelon' roses. *Postharvest Biology and Technology* **1**, 47-57.

van Doorn, W.G., De Strigter, H.C.M., De Witte, Y. and Boekestein, A. (1991c) Microorganisms at the cut surface and in xylem vessels of rose stems: a scanning

electron microscope study. *Journal of Applied Bacteriology* **70**, 34-39.

van Doorn, W.G., Zagory, D. and Reid. M.S. (1991d) Role of ethylene in vascular blockage in cut fronds from the fern *Adiantum raddianum*. *Scientia Horticulturae* **46**, 161-169.

van Doorn, W.G., Harkema, H. and Otma, E. (1991e) Is vascular blockage in stems of cut lilac flowers mediated by ethylene? *Acta Horticulturae* **298**, 177-181.

van Doorn, W.G., De Witte, Y. and Perik, R.R.J. (1990) Effect of antimicrobial compounds on the number of bacteria in stems of cut rose flowers. *Journal of Applied Bacteriology* **68**, 117-122.

van Doorn, W.G., Harkema, H. and Song, J.S. (1995) Water relations and senescence of cut *Iris* flowers: effects of cycloheximide. *Postharvest Biology and Technology* **5**, 345-351.

van Doorn, W.G., Schurer, K. and De Witte, Y. (1989) Role of endogenous bacteria in vascular blockage of cut rose flowers. *Plant Physiology* **134**, 375-381.

van Gorsel, R. (1993) Postharvest quality and water relations of Bouvardia cut flowers as affected by preharvest temperature and season. *Acta Horticulturae* **343**, 309-312.

van Kooten, O., Mensink, M.G.J., Otma, E.C., van Schaik, A.C.R. and Schouten, S.P. (1992) Chilling damage of dark stored cucumbers (*Cucumis sativus* L.) affects the maximum quantum yield of photosystem 2. p. 161-164. Progress in Photosynthesis Research, vol. IV. Kluwer Academic, Dordrecht, The Netherlands.

van Meeteren, U. (1978) Water relations and keeping-quality of cut gerbera flowers. I. The cause of stem break. *Scientia Horticulturae* **8**, 65-74.

van Meeteren, U. and van Gelder, H. (1999) Effect of time since harvest and handling conditions on rehydration ability of cut chrysanthemum flowers. *Postharvest Biology*

and Technology **16**, 169-177.

van Reinhold, N. (1991) Abscisic acid. In: Post-harvest physiology of perishable plant products, New York. 242-248 .

Vardi, Y. and Mayak, S. (1989) Involvement of abscisic acid during water stress and recovery in petunia flowers. *Acta Horticulturae* **261**, 107-112.

Venkatarayappa, T., Murrr, D.P. and Tsujita, M.J. (1981) Effect of Co^{2+} and sucrose on the physiology of cut Samantha roses. *Journal of Horticultural Science* **56**, 21-25.

Wahome, P.K., Jesch, H.H. and Grittner, I. (2001) Mechanisms of salt stress tolerance in two rose rootstocks: *Rosa chinensis* 'Major' and *R. rubiginosa*. *Scientia Horticulturae* **87**, 207-216.

Walker, M.A., Smith, D.M., Pauls, K.P. and McKersie, B.D. (1990) A chlorophyll fluorescence screening test to evaluate chilling tolerance in tomato. *HortScience* **25**, 334-339.

Wang, W.Y., Chen, W.S., Chen, W.H., Hung, L.S. and Chang, P.S. (2002) Influence of abscisic acid on flowering in *Phalaenopsis hybrida*. *Plant Physiology and Biochemistry* 40, 97-100.

Wilkinson, S. (1999) PH as a stress signal. *Plant Growth Regulation* **29**, 87-99.

Wilkinson, S. and Davis, W.J. (1997) Xylem sap pH increase: a drought signal received at the apoplastic face of the guard cell which involves the suppression of saturable ABA uptake by the epidermal symplast. *Plant Physiology* **113**, 559-573.

Wilkinson, S. and Davis, W.J. (2002) ABA-based chemical signalling: the co-ordination of responses to stress in plants. *Plant, Call and Environment* **25**, 195-210.

Wilkinson, S., Corlett, J.E., Oger, L. and Davies, W.J. (1998) Effects of xylem pH on transpiration from wild-type and *flacca* tomato leaves. *Plant Physiology* **117**, 703-

709.

Wilkinson, S., Clephan, A.L. Davies, W.J. (2001) Rapid low temperature-induced stomatal closure occurs in cold-tolerant *Commelina communis* leaves but not in cold-sensitive tobacco leaves, via a mechanism that involves apoplastic calcium but not abscisic acid. *Plant Physiology* **126**, 1566-1578.

Wilson, J.M. (1976) The mechanism of chill- and drought-hardening of *Pheseolus vulgaris* leaves. *New Phytologist* **76**, 257-270.

Wise, R.R. (1995) Chilling-enhanced photooxidation: the production, action and study of reactive oxygen species produced during chilling in the light. *Photosynthesis Research* **45**, 79-97.

Wittwer, S.H. (1983) Rising atmospheric CO_2 and crop productivity. *HortScience* **18**, 3-9.

Woltering, E.J. (1987) The effects of leakage of substances from mechanically wounded rose stems on bacterial growth and flower quality. *Scientia Horticulturae* **33**, 129-136.

Yamaguchi, H. and Hirata, Y. (1998) Influence of high temperature on flower stem length and photosynthesis of roses. *Acta Horticulturae* **454**, 391-393.

Zhong, R., Ripperger, A. and Ye, Z.H. (2000) Ectopic deposition of lignin in the pith of stems of two Arabidopsis mutants. *Plant Physiology* **123**, 59-70.

Zhuang, H., Hildebrand, D.F. and Barth, M.M. (1995) Senescence of Broccoli buds is related to changes in lipid peroxidation. *Journal of Agricultural Food Chemistry* **43**, 2585-2591.

Zieslin, N., Kohl, H.C., Kofranek, A.M., and Halevy, A.H. (1978) Changes in the water status of cut roses and its relationships to bent-neck phenomenon. *Journal of American Society for Horticultural Science* **103**, 176-179.

Zieslin, N. and Mor, Y. (1990) Light on Roses. A review. *Scientia Horticulturae* **43**, 1-4.

APPENDIX 1
GENERAL INTRODUCTION

Table A1.1: Main importing countries for horticultural products in 2001-2003.

Country	Year		
	2001	2002	2003
Germany	1496	1512	1585
United Kingdom	652	729	741
France	594	606	649
Italy	257	281	317
Belgium	147	164	184
Switzerland	128	141	134
Austria	121	127	133
Denmark	99	107	123
United States of America	144	143	115
Russia	74	87	104
Other countries	600	671	678

Source: Dutch Floricultural Wholesale Board.

Table A1.2: Top 10 flower species sold at The Netherlands auctions in 2001-2003 in 10^6 single flower stems. Source: The Federation of Netherlands Flower Auctions, 2004.

Product	Year		
	2001	2002	2003
Rose	1685	1740	1854
Tulip	543	600	628
Spray chrysanthemum	448	474	467
Gerbera (Transvaal daisy)	294	295	298
Lily	157	157	152
Freesia	132	133	132
Alstroemeria (Jersey lily)	143	131	128
Hypericum	72	73	80
Gypsophila (Baby's breath)	82	82	80
Iris	89	76	72

APPENDIX 2
LITERATURE REVIEW

Table A2.1: Optimal development stage of different cut flower species harvested for direct sale (after Nowak and Rudnicki, 1990).

Botanical name	Stage of development
Acacia sp.	1/2 florets open
Achillea filipendulina	fully open flowers
Aconitum napellus	1/2 florets open
Agapanthus umbellatus	1/4 florets open
Allium giganteum	1/3 florets open
Allium sphaerocephalon	1/4 florets open
Alstromeria hybrids	4-5 florets open
Althea rosea	1/3 florets open
Amaranthus	1/2 florets open
Anemone coronaria	buds beginning to open
Anthurium sp.	spadix almost fully developed
Antirrhinum majus	1/3 florets open
Aquilegia hybrids	1/2 florets open
Astilbe hybrids	1/2 florets open
Bellis perennis	fully open flowers
Bouvardia hybrids	flowers beginning to open
Calendula officinalis	fully open flowers
Callistephus chinensis	fully open flowers
Camellia japonica	fully open flowers
Campanula sp.	1/2 florets open
Cattleya sp.	3-4 days after opening
Celosia argentea	1/2 florets open
Centaurea	flowers beginning to open
Cheiranthus cheirii	1/2 florets open
Chrysanthemum sp.	fully open flowers
Standard cultivars	outer petals fully elongated
Spray cultivars	outer petals fully elongated
Singles	open but before anthesis
Anemones	open but before disk flowers start to elongate
Pompons and decorative	centre of the oldest flower fully open
Clarkia elegans	1/2 florets open
Clivia miniata	1/4 florets open
Consolida ambigua	2-5 florets open
Convallaria majalis	1/2 florets open, terminal bud having lost green colour
Coreopsis grandiflora	fully open flowers
Costus sp.	almost fully open flowers
Crocosmia crocosmiflora	fully open flowers

Table A2.1: Continued.

Botanical name	Stage of development
Cyclamen persicum	fully open flowers
Cymbidium sp.	3-4 days after opening
Dahlia variabilis	fully open flowers
Delphinium sp.	1/2 florets open
Dendrobium sp.	almost fully open flowers
Dianthus sp.	1/2 florets open
Standard cultivars	half-open flowers
Spray cultivars	2 fully open flowers
Digitalis purpurea	1/2 florets open
Doronicum caucasicum	almost open flowers
Echinops ritro	half-open flowers
Eremurus robustus	1/2 florets open
Erica sp.	1/2 florets open
Erigeron hybrids	fully open flowers
Eryngium sp.	fully open flowers
Eucharis grandiflora	almost open flowers
Euphorbia fulgens	showing enough colour
Euphorbia pulcherrima	fully mature
Eustoma russellianum	5-6 open flowers
Freesia hybrids	first bud beginning to open
Fritillaria imperialis	half-open flowers
Gaillardia sp.	fully open flowers
Gardenia jasminoides	almost fully open flowers
Gerbera jamesonii	outer row of flowers showing pollen
Gladiolus sp.	1-5 buds showing colour
Gloriosa superba	almost fully open flowers
Gypsophila sp.	flowers open but not overly mature
Helianthus annus	fully open flowers
Heliopsis helianthoides	fully open flowers
Helleborus niger	half-open flowers
Hemerocallis sp.	half-open flowers
Hippeastrum hybrids	colored buds
Iris sp.	colored buds
Ixia sp.	colored buds
Kalanchoe hybrids	1/2 florets open
Kniphofia uvaria	almost all florets showing colour
Lathyrus odoratus	1/2 florets open
Leontopodium alpinum	fully open flowers
Liatris spicata	1/2 florets open
Lilium sp.	colored buds
Limonium sp.	almost fully open flowers
Lupinus sp.	1/2 florets open
Matthiola incana	1/2 florets open
Monarda didyma	almost open flowers

Table A2.1: Continued.

Botanical name	Stage of development
Muscari botryoides	1/2 florets open
Myosotis silvatica	1/2 florets open
Narcissus sp.	'goose neck' stage
Nepeta faassenii	1/2 florets open
Nerine bowdenii	oldest buds almost open
Nigella damascena	open flowers
Ornithogalum umbellatum	colored buds
Paeonia sp.	colored buds
Papaver sp.	colored buds
Paphiopedilum sp.	3-4 days after opening
Phalaenopsis sp.	3-4 days after opening
Phlox paniculata	½ florets open
Polianthes tuberosa	majority of florets open
Primula sp.	1/2 florets open
Ranunculus asiaticus	buds beginning to open
Reseda odorata	1/2 florets open
Rosa hybrids	
Red and pink cultivars	first 2 petals beginning to unfold, calyx reflexed below a horizontal position
Yellow cultivars	slightly earlier than red and pink
White cultivars	slightly later than red and pink
Rudbeckia sp.	fully open flowers
Scabiosa sp.	half-open flowers
Scilla sibirica	half-open flowers
Sedum sp.	fully open flowers
Solidago sp.	1/2 florets open
Stephanotis floribunda	fully open flowers
Strelitzia reginae	first floret open
Tagetes erecta	fully open flowers
Thalictrum aguilegifolium	1/2 florets open
Trollius sp.	half-open flowers
Tropaeolum majus	fully open flowers
Tulipa gesneriana	half-colored buds
Veronica sp.	1/2 florets open
Viola sp.	almost open flowers
Zantedeschia sp.	just before the spathe begins to turn downward
Zinnia elegans	fully open flowers

Table A2.2: Identification of bacteria from flower vase water, after several days of vase life, as related to the flower species. Data from van Doorn (1997).

Bacteria	Rose	Chrysanthemum	Gerbera
GRAM-NEGATIVE RODS			
Acinertobacter sp.		+	+
Achromobacter sp.	+		
Alcaligenes sp.	+		+
Citrobacter freundii	+		
C. freundii var. amalonaticus	+		+
Enterobacter			
E. agglomerans	+	+	+
E. cloacae	+	+	+
E. gergovinae		+	+
Enterobacter sp.	+		+
Flavobacterium sp.	+		
Pseudomonas aeruginosa	+		
P. aeruginosa	+	+	
P. capacia	+	+	
P. fluorescens	+		+
P. maltophilia	+	+	
P. mendocina	+	+	
P. pikettii	+		+
P. putida	+	+	+
P. putrefaciens	+		
P. stutzeri	+		
P. vesicularis	+	+	
Pseudomonas sp.	+		
GRAM-POSITIVE RODS			
Bacillus cereus (lecithinase -)	+		+
B. cereus (lecithinase +)	+	+	
B. circulans			+
B. licheniformis	+		
B. mycoides		+	+
B. polymyxa	+	+	+
B. subtilis	+	+	+
B. subtilis var. niger		+	+
B. thiaminolyticus			+
Corynebacteria	+		
GRAM-POSITIVE COCCI			
Streptococcus lactis group		+	+

Note. + = positive identification.

Table A2.3: Identification of filamentous fungi and yeast in the vase water of some cut flowers after 3-12 days and Ruscus cut foliage after 30 days of vase life. The Central Bureau for Fungal Cultures (Baarn, Holland) made all identifications except those on Ruscus. Data from van Doorn (1997).

Fungus	Rose	Chrysanthemum	Gerbera	Ruscus
Aspergillus niger				+
A. terreus				+
Aureobasidium pullulans (yeast-like fungus)			+	
Botrytis cinerea			+	
Botrytis sp.	+			
Cladosporium herbicola				+
Fusarium solani		+		
F. oxysporum	+		+	+
Mucor hiemalis	+	+	+	
M. racemosus			+	
Penicillium brevicompactum			+	
Penicillium sp.				+
Rhizopus stolonifer	+	+	+	
Rhizopus sp.	+			
Trichoderma pseudokoningii		+		
Verticillium brevicompactum	+			

Note. + = positive identification.

Table A2.4: Examples of flowers and cut greens in which antimicrobial compounds had a positive effect on the length of vase life. The compounds either included in the vase solution at the beginning of vase life at about 20°C, or were given as a pulse treatment immediately after harvest, at various periods and temperatures, which are indicated in parentheses. Pulse treatments temperatures were about 20°C unless mentioned otherwise (after van Doorn, 1997).

Compound	Species	Concentration (mg l^{-1})	Reference
METAL SALTS			
$Al(SO_4)_2$			
	Forsythia	800	A. Ruting, pers. Comm., 1995
	Lupinus hartwegii	100-300	Mohan Ram and Rao 1977
	Phalaenopsis	800	A. Ruting, pers. Comm.
	Rosa hybrida	400-800	van Doorn, 1997
$Co(CH_3COO)_2$	*R. hybrida*	266	Venkatarayappa et al. 1981
$CoCl_2$	*R. hybrida*	260	Venkatarayappa et al. 1981; Reddy 1988
	Tagetes patula	13-65	Chandra et al. 1981
$Co(NO_3)_2$	*Adiantum radianum*	185-290	van Doorn et al. 1991a
	R. hybrida	275	Murr et al. 1979; Reddy 1988
$CoSO_4$	*R. hybrida*	132-310	Venkatarayappa et al. 1981; Reddy 1988
$NiCl_2$	*Phalaenopsis*	1500 (10 min)	van Doorn, 1997
$NiSO_4$	*R. hybrida*	1548 (10-20 min)	Reddy et al. 1988
$Ag(CH_3COO)_2$	*R. hybrida*	10-100	Ryan 1957; Scholes and Boodley 1964
$AgNO_3$	*Addiantum radianum* (fern)	12.5-25	van Doorn et al. 1991a
		1000 (20 min)	Fujino et al. 1983
	Antirrhinum majus	1000 (10-40 min)	Awad et al. 1986
	Argyranthemum frutescens	25	van Doorn, 1997
	Calendula	1000 (10-40 min)	Awad et al. 1986

Table A2.4: Continued.

Compound	Species	Concentration (mg l^{-1})	Reference
	Callistephus chinensis	1000 (10 min)	Kofranek et al. 1978
	Chrysanthemum	30 (24 h)	Nichols 1975
	Dendrobium	30	Ketsa and Boonrote 1990
	Gerbera jamesonii	20-30; 30 (20-24 h, 12 °C)	van Doorn, 1997
	Leptospermum	30	Joyce et al. 1993
	Polystichum (fern)	25	van Doorn, 1997
	R. hybrida	30-50	Scholes and Boodley 1964
		170-340 (30 min)	van Doorn et al. 1990
	Zinnia	1000 (10-40 min)	Awad et al. 1986
Zn(CH3COO)	*Rosa hybrida*	1-100	Ryan 1957
QUINOLINE COMPOUNDS			
HQC [A]	*Adiantum raddianum*	250-500	van Doorn et al. 1991d
	Gladiolus	450-600	van Doorn, 1997
	Gypsophila paniculata	250	Jones and Hill 1993
	R. hybrida	250	Burdett 1970; van Doorn et al. 1990
	Ruhmohra adiantiformis	800 (10 min)	van Doorn, 1997
	Scilla campanulata	250	Jones and Hill 1993
	Syringa vulgaris	400	van Doorn et al. 1991e
HQS [A]	*Anigozanthos*	Not reported	Faragher 1989
	Argyranthemum frutescens	300	van Doorn, 1997
	Chrysanthemum	200	Gay and Nichols 1977
	Dendrobium	100	Ketsa and Boonrote 1990
	Leptospermum	200	Joyce et al. 1993
	R. hybrida	200	Burdett 1970

Table A2.4: Continued.

Compound	Species	Concentration (mg l^{-1})	Reference
	Syringa vulgaris	300	Sytsema-Kalkman 1991
CHLORINE COMPOUNDS			
BCDMH [A]	*Eucalyptus* (cut foliage)	10	Joyce et al. 1993
	Gerbera jamesonii	12	Jones and Hill 1993
	Lilium parkmannii	12	as above
	R. hybrida	12	as above
	Scilla campanulata	12	as above
DDMH [A]	*Antirrhinum majus*	200-300	Marousky 1976
	Argyranthemum fruescens	50	van Doorn, 1997
	Gladiolus	50-300	Marousky 1976
	Gypsophila paniculata	50	as above
	R. hybrida	50	as above
Chlorine bleach	*R. hybrida*	20-40	Ryan 1957
	Adiantum raddianum (fern)	10-20	van Doorn et al. 1991d
	Gerbera jamesonii	7.5	van Doorn, 1997
		40 (24 h)	J. N. van der Sprong, pers. Comm.
DICA	*Antirrhinum majus*	100-300	Kofranek et al. 1974; Marousky 1976
	Argyranthemum fruescens	200	Kofranek et al. 1974
	Aster	200	as above
	Dianthus caryophyllus	50	Marousky 1976
	Gerbera jamesonii	50	Jones and Hill 1993
	Gladiolus	50	Marousky 1976
	Gypsophila paniculata	50	as above; Jones and Hill 1993
	Lilium parkmanni	50	Jones and Hill 1993
	R. hybrida	50	Marousky 1976; Van Doorn et al. 1990

Table A2.4: Continued.

Compound	Species	Concentration (mg l^{-1})	Reference
	Scilla campanulata	50	Jones and Hill 1993
	Telopea speciosissima	25 available chlorine	Faragher 1986
	Thryptomene calycina	50	Jones et al. 1993
QUATERNARY AMMONIUM COMPOUNDS			
Physan	*Callistephus chinensis*	200	Kofranek et al. 1978
CHLORINATED HYDROCARBONS			
Chlorhexidine	*Aster*		van Doorn, 1997
Dichlorophen	*Gerbera jamesonii*		van Meeteren 1978
Hexachlorophen	*Aster*		van Doorn, 1997

Note [A]. BCDMH = 1-bromo-3-chloro-5.5-dimethylhydantoin; DDMH = 1.3-dichloro-5.5-dimethylhydantoin; DICA = dichlorocyanuric acid = sodium dichloro-s-triazone trione (SDT); HQC = 8-hydroxyquinoline citrate; HQS = 8-hydroxyquinoline sulphate; Physan = mixture of dimethylbenzylammonium chlorides and dimethylethylbenzylammonium chlorides.

APPENDIX 3
MATERIALS AND METHODS

Table A3.1: Mean concentration of the main nutritional elements in the growth media as calculated for central and eastern greenhouse from autumn 2002 to summer 2003. Concentrations were calculated after dilutions of used fertilisers.

Greenhouse	Concentration of Nutritional Elements (mg l^{-1})						E.C.	pH
	N	P	K^+	Ca^{2+}	Mg^{2+}	S	(mS cm^{-1})	
North	20.5	4.6	28.2	10.0	1.2	2.5	3.5-4.0	6.5-7.0
South-eastern	15.5	3.9	17.4	10.2	1.5	2.4	2.2-2.4	5.5-6.0

Note. The elements Mn^{2+}, Mo, B, Cu^{2+}, Zn^{2+} and Fe^{2+} were all at < 0.1 mg l^{-1}. E.C.2.2

Table A3.2: Determination of required dark adaptation time for 'First Red' and 'Akito' roses measuring the chlorophyll fluorescence ratio (F_v/F_m) at 2-minute intervals. Data are $\bar{x}$ ± SE; n=3. Bold numbers indicate the highest F_v/F_m value for 'First Red' and 'Akito' roses, respectively.

Time intervals (min)	Chlorophyll fluorescence (F_v/F_m)	
	'First Red'	'Akito'
2.0	0.695 (±0.01)	0.790 (±0.02)
4.0	0.793 (±0.02)	0.795 (±0.06)
6.0	0.800 (±0.11)	0.807 (±0.03)
8.0	0.810 (±0.01)	0.805 (±0.01)
10.0	0.815 (±0.03)	0.810 (±0.01)
12.0	0.823 (±0.01)	0.810 (±0.02)
14.0	0.828 (±0.03)	0.808 (±0.03)
16.0	**0.830 (±0.00)**	0.811 (±0.05)
18.0	0.825 (±0.07)	**0.823 (±0.05)**
20.0	0.827 (±0.03)	0.811 (±0.02)
22.0	0.828 (±0.03)	0.811 (±0.01)
24.0	0.810 (±0.01)	0.803 (±0.06)
26.0	0.812 (±0.08)	0.805 (±0.05)
28.0	0.806 (±0.03)	0.803 (±0.03)
30.0	0.803 (±0.02)	0.808 (±0.02)

Table A3.3: Determination of the appropriate saturating light intensity for 'First Red' and 'Akito' roses measuring the chlorophyll fluorescence ratio (F_v/F_m) after at least 15 minutes dark adaptation period. Data are $\bar{x} \pm$ SE; n=3. Bold numbers indicate the highest F_v/F_m value for 'First Red' and 'Akito' roses, respectively.

Light intensity	**Chlorophyll fluorescence (F_v/F_m)**	
(µmol m^{-2} s^{-1})	**'First Red'**	**'Akito'**
500	0.680 (±0.07)	0.702 (±0.07)
1000	0.775 (±0.04)	0.790 (±0.06)
1500	0.801 (±0.06)	0.810 (±0.04)
2000	**0.825 (±0.05)**	**0.815 (±0.09)**
2500	0.800 (±0.09)	0.810 (±0.09)
3000	0.792 (±0.07)	0.780 (±0.08)

Table A3.4: Retention times (min) and peak areas (mV*min) for ABA standard quantification by HPLC. Data are $\bar{x} \pm$ SE; n=3.

ABA standard (ng μl^{-1})	**Retention time (min)**	**Peak area (mV*min)**
100	12.37 (±0.20)	84.72 (±1.46)
10	14.38 (±0.35)	8.87 (±0.25)
1	14.69 (±0.26)	1.34 (±0.19)
0.1	13.04 (±0.25)	0.63 (±0.12)

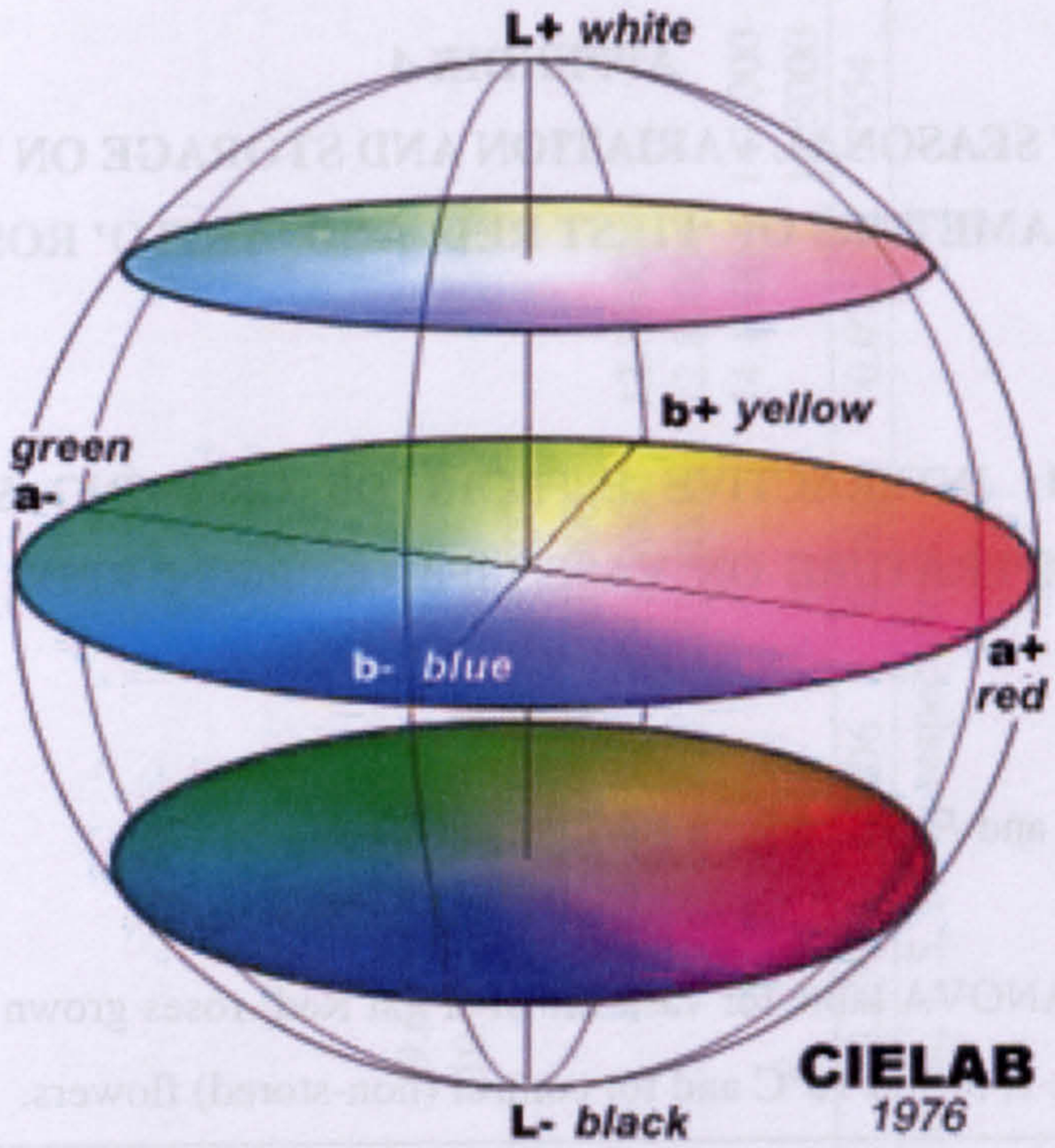


Plate A3.1: CIELAB (1976) a* and b* values are plotted on horizontal and vertical axes, respectively. L* indicates the lightness.

APPENDIX 4

EFFECT OF SEASONAL VARIATION AND STORAGE ON VASE LIFE PARAMETERS OF 'FIRST RED' AND 'AKITO' ROSES

APPENDIX 4.1: INTERACTIVE EFFECTS OF GROWING SEASON AND STORAGE TEMPERATURE ON VASE LIFE OF 'FIRST RED' AND 'AKITO' ROSES

A4.1.1: Vase life and F_v/F_m

Table A4.1.1.1: ANOVA table for vase life of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	325.084	3	108.361	95.922	0.000
Storage treatment (B)	693.409	3	231.136	204.602	0.000
AxB	244.328	9	27.148	24.031	0.000
Error	72.300	64	1.130		
Total	1335.122	79			

Table A4.1.1.2: ANOVA table for F_v/F_m on d 0 of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.074	3	0.024	18.393	0.000
Storage treatment (B)	0.037	3	0.012	9.240	0.000
AxB	0.046	9	0.005	3.839	0.001
Error	0.086	64	0.001		
Total	0.246	79			

Table A4.1.1.3: Means of vase life for 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6	7	8	9
winter10 [b]	20	2.9000								
winter1	20	3.7000	3.7000							
autumn10	20	3.8000	3.8000							
winter5	20		4.5000	4.5000						
summer10	20		4.6000	4.6000						
autumn5	20			5.8000						
autumn1	20			5.9000						
spring10	20				8.8000					
spring1	20				9.6000	9.6000				
spring5	20					10.7000	10.7000			
springC	20						11.5000	11.5000		
summer5	20						11.9000	11.9000		
winterC	20							12.8000	12.8000	
summer1	20							12.8000	12.8000	
autumnC	20								14.1000	14.1000
summerC	20									14.5000
Sig.		0.212	0.229	0.060	0.238	0.107	0.096	0.081	0.071	0.554

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.1.3: Means of vase life for 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6	7	8	9
winter10 [b]	20	2.9000								
winter1	20	3.7000	3.7000							
autumn10	20	3.8000	3.8000							
winter5	20		4.5000	4.5000						
summer10	20		4.6000	4.6000						
autumn5	20			5.8000						
autumn1	20			5.9000						
spring10	20				8.8000					
spring1	20				9.6000	9.6000				
spring5	20					10.7000	10.7000			
springC	20						11.5000	11.5000		
summer5	20						11.9000	11.9000		
winterC	20							12.8000	12.8000	
summer1	20							12.8000	12.8000	
autumnC	20								14.1000	14.1000
summerC	20									14.5000
Sig.		0.212	0.229	0.060	0.238	0.107	0.096	0.081	0.071	0.554

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.1.4: Means of F_v/F_m on d 0 for 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
winter1 [b]	20	0.6590			
winter5	20		0.7575		
winter10	20		0.7649	0.7649	
autumn1	20		0.8061	0.8061	0.8061
spring1	20			0.8114	0.8114
summer1	20			0.8114	0.8114
autumn5	20				0.8252
spring10	20				0.8264
summer10	20				0.8264
summerC	20				0.8267
autumnC	20				0.8274
autumn10	20				0.8325
springC	20				0.8349
spring5	20				0.8352
summer5	20				0.8352
winterC	20				0.8358
Sig.		1.000	0.052	0.072	0.300

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.1.5: ANOVA table for vase life of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	209.134	3	69.711	26.198	0.000
Storage treatment (B)	528.934	3	176.311	66.259	0.000
AxB	247.853	9	27.539	10.349	0.000
Error	170.300	64	2.661		
Total	1156.222	79			

Table A4.1.1.6: ANOVA table for F_v/F_m on d 0 of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.243	3	0.081	12.118	0.000
Storage treatment (B)	0.142	3	0.047	7.102	0.000
AxB	0.156	9	0.017	2.593	0.013
Error	0.428	64	0.006		
Total	0.969	79			

Table A4.1.1.7: Means of vase life for 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6	7	8	9
winter10 [b]	20	2.9000								
winter1	20	3.7000	3.7000							
autumn10	20	3.8000	3.8000							
winter5	20		4.5000	4.5000						
summer10	20		4.6000	4.6000						
autumn5	20			5.8000						
autumn1	20			5.9000						
spring10	20				8.8000					
spring1	20				9.6000	9.6000				
spring5	20					10.7000	10.7000			
springC	20						11.5000	11.5000		
summer5	20						11.9000	11.9000		
winterC	20							12.8000	12.8000	
summer1	20							12.8000	12.8000	
autumnC	20								14.1000	14.1000
summerC	20									14.5000
Sig.		0.212	0.229	0.060	0.238	0.107	0.096	0.081	0.071	0.554

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.1.8: Means of F_v/F_m on d 0 for 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
winter1 [b]	20	0.6590			
winter5	20		0.7575		
winter10	20		0.7649	0.7649	
autumn1	20		0.8061	0.8061	0.8061
spring1	20			0.8114	0.8114
summer1	20			0.8114	0.8114
autumn5	20				0.8252
spring10	20				0.8264
summer10	20				0.8264
summerC	20				0.8267
autumnC	20				0.8274
autumn10	20				0.8325
springC	20				0.8349
spring5	20				0.8352
summer5	20				0.8352
winterC	20				0.8358
Sig.		1.000	0.052	0.072	0.300

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.1.9: One-way ANOVA table for vase life of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	9.050	3	3.017	3.742	0.033
Within Groups	12.900	16	0.806		
Total	21.950	19			

Table A4.1.1.10: One-way ANOVA table for vase life of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	58.550	3	19.517	11.828	0.000
Within Groups	26.400	16	1.650		
Total	84.950	19			

Table A4.1.1.11: One-way ANOVA table for vase life of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	103.450	3	34.483	11.591	0.000
Within Groups	47.600	16	2.975		
Total	151.050	19			

Table A4.1.1.12: One-way ANOVA table for vase life of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	111.738	3	37.246	21.909	0.000
Within Groups	27.200	16	1.700		
Total	138.938	19			

Table A4.1.1.13: One-way ANOVA table for vase life of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.200	3	1.400	0.563	0.647
Within Groups	39.800	16	2.488		
Total	44.000	19			

Table A4.1.1.14: One-way ANOVA table for vase life of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	29.700	3	9.900	3.300	0.047
Within Groups	48.000	16	3.000		
Total	77.700	19			

Table A4.1.1.15: One-way ANOVA table for vase life of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	122.500	3	40.833	42.982	0.000
Within Groups	15.200	16	0.950		
Total	137.700	19			

Table A4.1.1.16: One-way ANOVA table for vase life of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	97.838	3	32.613	5.811	0.007
Within Groups	89.800	16	5.613		
Total	187.638	19			

Table A4.1.1.17: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0022	3	0.00075	37.583	0.000
Within Groups	0.0003	16	0.00002		
Total	0.0025	19			

Table A4.1.1.18: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0018	3	0.00060	34.571	0000
Within Groups	0.0002	16	0.00001		
Total	0.0020	19			

Table A4.1.1.19: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0020	3	0.0006	2.563	0.091
Within Groups	0.0043	16	0.0002		
Total	0.0064	19			

Table A4.1.1.20: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.078	3	0.026	5.088	0.012
Within Groups	0.082	16	0.005		
Total	0.161	19			

Table A4.1.1.21: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.200	3	1.400	0.563	0.647
Within Groups	39.800	16	2.488		
Total	44.000	19			

Table A4.1.1.22: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	29.700	3	9.900	3.300	0.047
Within Groups	48.000	16	3.000		
Total	77.700	19			

Table A4.1.1.23: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	122.500	3	40.833	42.982	0.000
Within Groups	15.200	16	0.950		
Total	137.700	19			

Table A4.1.1.24: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	97.838	3	32.613	5.811	0.007
Within Groups	89.800	16	5.613		
Total	187.638	19			

Table A4.1.1.25: Parameters estimated for the linear model ($y = y_0 +/- ax$) used to describe the effects of storage temperature (1, 5, and 10°C) on mean vase life and F_v/F_m on d 0 of 'First Red' and 'Akito' roses.

Temperature (°C)	Estimated parameters		Coefficient (R^2)
	y_0	a	
a. Vase life			
'First Red'	8.91	-0.34	0.75
'Akito'	8.44	-0.07	0.88
b. F_v/F_m			
'First Red'	0.77	0.004	0.67
'Akito'	0.71	0.008	0.97

Table A4.1.1.26: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.079	3	0.026	5.139	0.011
Within Groups	0.082	16	0.005		
Total	0.161	19			

Table A4.1.1.27: One-way ANOVA table for F_v/F_m on d 4 of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.038	3	0.012	0.847	0.488
Within Groups	0.239	16	0.014		
Total	0.277	19			

Table A4.1.1.28: One-way ANOVA table for F_v/F_m on d 8 of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.038	3	0.012	0.954	0.438
Within Groups	0.214	16	0.013		
Total	0.252	19			

Table A4.1.1.29: One-way ANOVA table for F_v/F_m on d 12 of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.041	3	0.013	1.109	0.375
Within Groups	0.199	16	0.012		
Total	0.241	19			

Table A4.1.1.30: One-way ANOVA table for F_v/F_m on d 16 of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.028	3	0.009	0.405	0.752
Within Groups	0.374	16	0.023		
Total	0.403	19			

Table A4.1.1.31: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.262	3	0.087	5.060	0.012
Within Groups	0.276	16	0.017		
Total	0.538	19			

Table A4.1.1.32: One-way ANOVA table for F_v/F_m on d 4 of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.203	3	0.067	4.524	0.018
Within Groups	0.239	16	0.014		
Total	0.442	19			

Table A4.1.1.33: One-way ANOVA table for F_v/F_m on d 8 of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.198	3	0.065	4.575	0.017
Within Groups	0.230	16	0.014		
Total	0.428	19			

Table A4.1.1.34: One-way ANOVA table for F_v/F_m on d 12 of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.147	3	0.048	2.281	0.118
Within Groups	0.343	16	0.021		
Total	0.490	19			

Table A4.1.1.35: One-way ANOVA table for F_v/F_m on d 16 of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.157	3	0.052	2.541	0.093
Within Groups	0.329	16	0.020		
Total	0.486	19			

A4.1.2: Flower and foliage stages

Table A4.1.2.1: ANOVA table for flower stages (d 8) of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	5.463	3	1.821	8.828	0.000
Storage treatment (B)	13.163	3	4.388	21.273	0.000
AxB	3.663	9	0.407	1.973	0.057
Error	13.200	64	0.206		
Total	35.488	79	35.488		

Table A4.1.2.2: Non-parametric test (Kruskal-Wallis) for flower stages (score 1-5) of 'First Red' roses grown during the year.

Growing seasons	N		Mean Rank
autumn	80		42.65
winter	80		32.53
spring	80		52.83
summer	80		34.00
Total	20		
Variable	d.f.	Chi-square	Asymp. Sig.
Flower stages (day 8)	3	10.274	0.016

Table A4.1.2.3: Non-parametric test (Kruskal-Wallis) for flower stages (score 1-5) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) flowers.

Storage treatment	N		Mean Rank
control	80		63.00
1°C	80		23.33
5°C	80		36.75
10°C	80		38.92
Total	20		
Variable	d.f.	Chi-square	Asymp. Sig.
Flower stages (day 8)	3	32.023	0.000

Table A4.1.2.4: ANOVA table for flower stages (d 8) of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	32.109	3	10.703	32.775	0.000
Storage treatment (B)	16.509	3	5.503	16.852	0.000
AxB	6.328	9	0.703	2.153	0.037
Error	20.900	64	0.327		
Total	75.847	79			

Table A4.1.2.5: Non-parametric test (Kruskal-Wallis) for flower stages (score 1-5) of 'Akito' roses grown during the year.

Growing seasons	N	Mean Rank
autumn	80	39.40
winter	80	16.42
spring	80	54.03
summer	80	52.15
Total	20	

Variable	d.f.	Chi-square	Asymp. Sig.
Flower stages (day 8)	3	34.036	0.000

Table A4.1.2.6: Non-parametric test (Kruskal-Wallis) for flower stages (score 1-5) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) flowers.

Storage treatment	N	Mean Rank
control	80	56.03
1°C	80	26.55
5°C	80	38.08
10°C	80	41.35
Total	20	

Variable	d.f.	Chi-square	Asymp. Sig.
Flower stages (day 8)	3	16.734	0.001

Table A4.1.2.7: Means of flower stage (d 8) for 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5
summer1 [b]	20	1.8000				
winter10	20	1.9000	1.9000			
winter1	20	2.0000	2.0000			
spring1	20	2.0000	2.0000			
summer5	20	2.0000	2.0000			
winter5	20	2.1000	2.1000			
autumn1	20	2.3000	2.3000	2.3000		
autumn10	20	2.3000	2.3000	2.3000		
autumn5	20		2.5000	2.5000	2.5000	
summer10	20		2.5000	2.5000	2.5000	
summerC	20			2.9000	2.9000	2.9000
spring5	20				3.0000	3.0000
autumnC	20				3.1000	3.1000
winterC	20				3.1000	3.1000
spring10	20					3.2000
springC	20					3.5000
Sig.		0.143	0.081	0.066	0.070	0.070

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.2.8: Means of flower stage (d 8) for 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5
summer1 [b]	20	1.9000				
winter10	20	2.0000	2.0000			
winter1	20	2.1000	2.1000			
spring1	20		2.7000	2.7000		
summer5	20			3.0000	3.0000	
winter5	20			3.0000	3.0000	
autumn1	20			3.1000	3.1000	
autumn10	20			3.1000	3.1000	
autumn5	20			3.2000	3.2000	
summer10	20				3.6000	3.6000
summerC	20				3.7000	3.7000
spring5	20					4.0000
autumnC	20					4.1000
winterC	20					4.1000
spring10	20					4.3000
springC	20					
Sig.		0.606	0.071	0.234	0.099	0.094

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.2.9: ANOVA table for foliage stages (d 8) of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.659	3	0.220	1.234	0.305
Storage treatment (B)	11.259	3	3.753	21.070	0.000
AxB	4.353	9	0.484	2.715	0.010
Error	11.400	64	0.178		
Total	27.672	79			

Table A4.1.2.10: Non-parametric test (Kruskal-Wallis) for foliage stages (score 1-5) of 'First Red' roses grown during the year.

Growing seasons	N		Mean Rank
autumn	80		40.50
winter	80		39.50
spring	80		35.85
summer	80		46.15
Total	20		
Variable	d.f.	Chi-square	Asymp. Sig.
Foliage stages (day 8)	3	2.151	0.542

Table A4.1.2.11: Non-parametric test (Kruskal-Wallis) for foliage stages (score 1-5) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) flowers.

Storage treatment	N	Mean Rank
control	80	16.40
1°C	80	48.10
5°C	80	44.50
10°C	80	53.00
Total	20	

Variable	d.f.	Chi-square	Asymp. Sig.
Foliage stages (day 8)	3	31.978	0.000

Table A4.1.2.12: ANOVA table for foliage stages (d 8) of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	1.038	3	0.346	1.203	0.316
Storage treatment (B)	2.312	3	0.771	2.681	0.054
AxB	3.238	9	0.360	1.251	0.281
Error	18.400	64	0.287		
Total	24.987	79			

Table A4.1.2.13: Non-parametric test (Kruskal-Wallis) for foliage stages (score 1-5) of 'Akito' roses grown during the year.

Growing seasons	N	Mean Rank
autumn	80	39.45
winter	80	36.17
spring	80	47.90
summer	80	38.47
Total	20	

Variable	d.f.	Chi-square	Asymp. Sig.
Foliage stages (day 8)	3	3.110	0.375

Table A4.1.2.14: Non-parametric test (Kruskal-Wallis) for foliage stages (score 1-5) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) flowers.

Storage treatment	N	Mean Rank
control	80	29.10
1°C	80	46.95
5°C	80	39.08
10°C	80	46.88
Total	20	

Variable	d.f.	Chi-square	Asymp. Sig.
Foliage stages (day 8)	3	8.469	0.037

Table A4.1.2.15: Means of foliage stage (d 8) for 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5
summer1 [b]	20	2.6000				
winter10	20	2.9000	2.9000			
winter1	20	3.0000	3.0000			
spring1	20	3.1000	3.1000			
summer5	20		3.2000	3.2000		
winter5	20		3.4000	3.4000	3.4000	
autumn1	20		3.4000	3.4000	3.4000	
autumn10	20		3.5000	3.5000	3.5000	
autumn5	20			3.8000	3.8000	3.8000
summer10	20			3.8000	3.8000	3.8000
summerC	20			3.8000	3.8000	3.8000
spring5	20				3.9000	3.9000
autumnC	20				3.9000	3.9000
winterC	20				4.0000	4.0000
spring10	20				4.0000	4.0000
springC	20					4.2000
Sig.		0.091	0.054	0.054	0.062	0.208

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.2.16: One-way ANOVA table for flower stages of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	9.050	3	3.017	3.742	0.033
Within Groups	12.900	16	0.806		
Total	21.950	19			

Table A4.1.2.17: One-way ANOVA table for flower stages of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	58.550	3	19.517	11.828	0.000
Within Groups	26.400	16	1.650		
Total	84.950	19			

Table A4.1.2.18: One-way ANOVA table for flower stages of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	103.450	3	34.483	11.591	0.000
Within Groups	47.600	16	2.975		
Total	151.050	19			

Table A4.1.2.19: One-way ANOVA table for flower stages of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	111.738	3	37.246	21.909	0.000
Within Groups	27.200	16	1.700		
Total	138.938	19			

Table A4.1.2.20: One-way ANOVA table for flower stages of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.200	3	1.400	0.563	0.647
Within Groups	39.800	16	2.488		
Total	44.000	19			

Table A4.1.2.21: One-way ANOVA table for flower stages of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	29.700	3	9.900	3.300	0.047
Within Groups	48.000	16	3.000		
Total	77.700	19			

Table A4.1.2.22: One-way ANOVA table for flower stages of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	122.500	3	40.833	42.982	0.000
Within Groups	15.200	16	0.950		
Total	137.700	19			

Table A4.1.2.23: One-way ANOVA table for flower stages of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	97.838	3	32.613	5.811	0.007
Within Groups	89.800	16	5.613		
Total	187.638	19			

Table A4.1.2.24: One-way ANOVA table for foliage stage of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0022	3	0.00075	37.583	0.000
Within Groups	0.0003	16	0.00002		
Total	0.0025	19			

Table A4.1.2.25: One-way ANOVA table for foliage stage of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0018	3	0.00060	34.571	0000
Within Groups	0.0002	16	0.00001		
Total	0.0020	19			

Table A4.1.2.26: One-way ANOVA table for foliage stage of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0020	3	0.0006	2.563	0.091
Within Groups	0.0043	16	0.0002		
Total	0.0064	19			

Table A4.1.2.27: One-way ANOVA table for foliage stage of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.078	3	0.026	5.088	0.012
Within Groups	0.082	16	0.005		
Total	0.161	19			

Table A4.1.2.28: One-way ANOVA table for foliage stage of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.200	3	1.400	0.563	0.647
Within Groups	39.800	16	2.488		
Total	44.000	19			

Table A4.1.2.29: One-way ANOVA table for foliage stage of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	29.700	3	9.900	3.300	0.047
Within Groups	48.000	16	3.000		
Total	77.700	19			

Table A4.1.2.30: One-way ANOVA table for foliage stage of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	122.500	3	40.833	42.982	0.000
Within Groups	15.200	16	0.950		
Total	137.700	19			

Table A4.1.2.31: One-way ANOVA table for foliage stage of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	97.838	3	32.613	5.811	0.007
Within Groups	89.800	16	5.613		
Total	187.638	19			

A4.1.3: Flower and foliage stages during vase life

Table 4.1.3.1: One-way ANOVA table for flower stage (day 0) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.494	3	0.831	34.885	0.000
Within Groups	0.381	16	0.023		
Total	2.875	19			

Table 4.1.3.2: One-way ANOVA table for flower stage (day 4) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.359	3	0.453	10.835	0.000
Within Groups	0.669	16	0.041		
Total	2.027	19			

Table 4.1.3.3: One-way ANOVA table for flower stage (day 8) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.291	3	1.097	17.772	0.000
Within Groups	0.988	16	0.061		
Total	4.278	19			

Table 4.1.3.4: One-way ANOVA table for flower stage (day 12) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.634	3	1.211	22.968	0.000
Within Groups	0.844	16	0.052		
Total	4.477	19			

Table 4.1.3.5: One-way ANOVA table for flower stage (day 16) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.606	3	1.202	21.981	0.000
Within Groups	0.875	16	0.054		
Total	4.481	19			

Table 4.1.3.6: One-way ANOVA table for flower stage (day 0) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.365	3	0.788	39.569	0.000
Within Groups	0.319	16	0.019		
Total	2.684	19			

Table 4.1.3.7: One-way ANOVA table for flower stage (day 4) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.237	3	0.746	16.319	0.000
Within Groups	0.731	16	0.045		
Total	2.969	19			

Table 4.1.3.8: One-way ANOVA table for flower stage (day 8) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.896	3	0.965	11.441	0.000
Within Groups	1.350	16	0.084		
Total	4.246	19			

Table 4.1.3.9: One-way ANOVA table for flower stage (day 12) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.234	3	1.078	10.615	0.000
Within Groups	1.625	16	0.102		
Total	4.859	19			

Table 4.1.3.10: One-way ANOVA table for flower stage (day 16) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.402	3	1.134	11.476	0.000
Within Groups	1.581	16	0.098		
Total	4.984	19			

Table 4.1.3.11: One-way ANOVA table for foliage stage (day 0) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	6.703	3	2.234	67.294	0.000
Within Groups	0.531	16	0.033		
Total	7.234	19			

Table 4.1.3.12: One-way ANOVA table for foliage stage (day 4) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.259	3	1.086	32.341	0.000
Within Groups	0.538	16	0.033		
Total	3.797	19			

Table 4.1.3.13: One-way ANOVA table for foliage stage (day 8) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.815	3	0.938	23.096	0.000
Within Groups	0.650	16	0.040		
Total	3.465	19			

Table 4.1.3.14: One-way ANOVA table for foliage stage (day 12) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.212	3	0.070	1.259	0.322
Within Groups	0.900	16	0.056		
Total	1.113	19			

Table 4.1.3.15: One-way ANOVA table for foliage stage (day 16) of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.102	3	0.034	0.355	0.786
Within Groups	1.537	16	0.096		
Total	1.640	19			

Table 4.1.3.16: One-way ANOVA table for foliage stage (day 0) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	8.277	3	2.759	103.873	0.000
Within Groups	0.425	16	0.026		
Total	8.702	19			

Table 4.1.3.17: One-way ANOVA table for foliage stage (day 4) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.603	3	0.534	13.818	0.000
Within Groups	0.619	16	0.038		
Total	2.222	19			

Table 4.1.3.18: One-way ANOVA table for foliage stage (day 8) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.578	3	0.193	2.442	0.102
Within Groups	1.263	16	0.078		
Total	1.841	19			

Table 4.1.3.19: One-way ANOVA table for foliage stage (day 12) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.194	3	0.064	1.704	0.206
Within Groups	0.606	16	0.037		
Total	0.800	19			

Table 4.1.3.20: One-way ANOVA table for foliage stage (day 16) of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.128	3	0.042	0.816	0.504
Within Groups	0.837	16	0.052		
Total	0.966	19			

A4.1.4: Petal colour

Table A4.1.4.1: Means of b* value (d 8) for 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6	7	8	9	10
autumn5 [b]	20	-0.6100									
winter10	20	0.5800	0.5800								
autumn1	20	1.3000	1.3000	1.3000							
winter5	20		3.7000	3.7000	3.7000						
autumn10	20			4.0400	4.0400	4.0400					
summer10	20				4.7660	4.7660					
winterC	20				6.5200	6.5200	6.5200				
winter1	20					7.0700	7.0700	7.0700			
autumnC	20						8.3100	8.3100	8.3100		
spring1	20							9.8500	9.8500	9.8500	
spring5	20								10.8900	10.8900	
summer1	20									12.0120	
spring10	20									12.1200	
summer5	20									12.1860	
springC	20									12.5100	
summerC	20										17.0210
Sig.		0.237	0.053	0.090	0.091	0.069	0.268	0.085	0.110	0.127	1.000

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.4.2: ANOVA table for b* value (d 8) of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	1156.566	3	385.522	68.018	0.000
Storage treatment (B)	364.821	3	121.607	21.455	0.000
AxB	398.067	9	44.230	7.804	0.000
Error	362.746	64	5.668		
Total	2282.201	79			

Table A4.1.4.3: One-way ANOVA table for b* value (d 8) of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	21.999	3	7.333	1.185	0.347
Within Groups	99.052	16	6.191		
Total	121.051	19			

Table A4.1.4.4: One-way ANOVA table for b* value (d 8) of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	382.804	3	127.601	77.942	0.000
Within Groups	26.194	16	1.637		
Total	408.998	19			

Table A4.1.4.5: One-way ANOVA table for b* value (d 8) of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	224.647	3	74.882	6.975	0.003
Within Groups	171.761	16	10.735		
Total	396.408	19			

Table A4.1.4.6: One-way ANOVA table for b* value (d 8) of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	133.437	3	44.479	10.826	0.000
Within Groups	65.739	16	4.109		
Total	199.176	19			

Table A4.1.4.7: One-way ANOVA table for b* value of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 0).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	61.177	3	20.392	14.865	0.000
Within Groups	21.949	16	1.372		
Total	83.127	19			

Table A4.1.4.8: One-way ANOVA table for b* value of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 4).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	98.159	3	32.720	39.064	0.000
Within Groups	13.402	16	0.838		
Total	111.561	19			

Table A4.1.4.9: One-way ANOVA table for b* value of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 8).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	91.205	3	30.402	26.814	0.000
Within Groups	18.141	16	1.134		
Total	109.346	19			

Table A4.1.4.10: One-way ANOVA table for b* value of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 12).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	38.663	3	12.888	8.288	0.001
Within Groups	24.881	16	1.555		
Total	63.544	19			

Table A4.1.4.11: One-way ANOVA table for b* value of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 16).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.961	3	1.654	1.015	0.412
Within Groups	26.074	16	1.630		
Total	31.035	19			

A4.1.5 Bent neck

Table A4.1.5.1: ANOVA table for bent neck incidence of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	375.000	3	125.000	1.000	0.399
Storage treatment (B)	375.000	3	125.000	1.000	0.399
AxB	1125.000	9	125.000	1.000	0.449
Error	8000.000	64	125.000		
Total	9875.000	79			

Table A4.1.5.2: ANOVA table for bent neck incidence of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	12000.000	3	4000.000	3.048	0.035
Storage treatment (B)	7000.000	3	2333.333	1.778	0.160
AxB	25000.000	9	2777.778	2.116	0.041
Error	84000.000	64	1312.500		
Total	128000.000	79			

A4.1.6: Solution usage and fresh weight

Table A4.1.6.1: ANOVA table for solution usage (d 7) by 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.479	3	0.160	1.691	0.178
Storage treatment (B)	0.491	3	0.164	1.737	0.168
AxB	1.372	9	0.152	1.616	0.130
Error	6.037	64	0.094		
Total	8.379	79			

Table A4.1.6.2: ANOVA table for solution usage (d 7) by 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	1.958	3	0.653	42.441	0.000
Storage treatment (B)	1.041	3	0.347	22.555	0.000
AxB	1.048	9	0.116	7.573	0.000
Error	0.984	64	0.015		
Total	5.032	79			

Table A4.1.6.3: Means of solution usage (d 7) by 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6	7
winter1 [b]	20	0.0442						
winter10	20	0.0511						
winter5	20	0.1211	0.1211					
summer10	20		0.2498	0.2498				
spring10	20			0.2880	0.2880			
summer5	20			0.3135	0.3135			
summerC	20			0.3329	0.3329			
summer1	20				0.4258	0.4258		
spring1	20				0.4490	0.4490	0.4490	
spring5	20				0.4616	0.4616	0.4616	
autumn5	20					0.5796	0.5796	0.5796
autumn10	20					0.5808	0.5808	0.5808
springC	20						0.6063	0.6063
autumn1	20							0.7225
winterC	20							0.7303
autumnC	20							0.7380
Sig.		0.361	0.106	0.342	0.055	0.082	0.077	0.081

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.6.4: ANOVA table for fresh weight (d 8) of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	4348.135	3	1449.378	29.776	0.000
Storage treatment (B)	2037.594	9	226.399	4.651	0.000
AxB	2037.594	9	226.399	4.651	0.000
Error	3115.293	64	48.676		
Total	22574.523	79			

Table A4.1.6.5: Means of fresh weight (d 8) of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6	7
winter10 [b]	20	43.322						
winter5	20	46.942						
winter1	20	49.483						
spring10	20		69.754					
autumn5	20		70.051					
winterC	20		72.536					
summer10	20		75.236	75.236				
autumn10	20		76.034	76.034	76.034			
autumn1	20		76.794	76.794	76.794	76.794		
summer1	20			83.655	83.655	83.655	83.655	
spring1	20			84.211	84.211	84.211	84.211	
spring5	20				85.411	85.411	85.411	
springC	20					86.319	86.319	
summerC	20						91.086	91.086
summer5	20						91.698	91.698
autumnC	20							96.304
Sig.		0.193	0.169	0.073	0.061	0.057	0.116	0.271

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.6.6: ANOVA table for fresh weight (d 8) of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	4048.662	3	1349.554	9.053	0.000
Storage treatment (B)	8920.915	3	2973.638	19.948	0.000
AxB	3318.184	9	368.687	2.473	0.017
Error	9540.589	64	149.072		
Total	25828.350	79			

Table A4.1.6.7: Means of fresh weight (d 8) of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6
winter10 [b]	20	57.101					
winter1	20	57.887	57.887				
spring10	20	69.754	69.754	69.754			
winter5	20		74.106	74.106	74.106		
summer10	20			75.236	75.236		
autumn10	20			79.953	79.953		
spring5	20			85.411	85.411	85.411	
autumn5	20			85.824	85.824	85.824	
autumn1	20				89.116	89.116	
springC	20				90.198	90.198	
summer1	20				90.814	90.814	
summer5	20				91.698	91.698	
spring1	20				91.965	91.965	
summerC	20					98.052	98.052
winterC	20					102.189	102.189
autumnC	20						110.320
Sig.		0.127	0.050	0.072	0.054	0.069	0.139

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.6.8: One-way ANOVA table for solution usage (d 7) by 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0001	3	0.00005	0.016	0.997
Within Groups	0.0509	16	0.00318		
Total	0.0511	19			

Table A4.1.6.9: One-way ANOVA table for solution usage (d 7) by 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.035	3	0.011	10.449	0.000
Within Groups	0.018	16	0.001		
Total	0.054	19			

Table A4.1.6.10: One-way ANOVA table for solution usage (d 7) by 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.020	3	0.340	0.926	0.451
Within Groups	5.872	16	0.367		
Total	6.892	19			

Table A4.1.6.11: One-way ANOVA table for solution usage (d 7) by 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.807	3	0.269	44.976	0.000
Within Groups	0.095	16	0.005		
Total	0.903	19			

Table A4.1.6.12: One-way ANOVA table for solution usage (d 7) by 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.254	3	0.084	24.940	0.000
Within Groups	0.054	16	0.003		
Total	0.308	19			

Table A4.1.6.13: One-way ANOVA table for solution usage (d 7) by 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.079	3	0.026	2.788	0.074
Within Groups	0.152	16	0.009		
Total	0.231	19			

Table A4.1.6.14: One-way ANOVA table for solution usage (d 7) by 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.113	3	0.037	0.931	0.448
Within Groups	0.648	16	0.040		
Total	0.761	19			

Table A4.1.6.15: One-way ANOVA table for solution usage (d 7) by 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.642	3	0.547	67.325	0.000
Within Groups	0.130	16	0.008		
Total	1.773	19			

Table A4.1.6.16: One-way ANOVA table for fresh weight (d 8) of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	885.211	3	295.070	5.570	0.008
Within Groups	847.539	16	52.971		
Total	1732.750	19			

Table A4.1.6.17: One-way ANOVA table for fresh weight (d 8) of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1507.236	3	502.412	33.953	0.000
Within Groups	236.760	16	14.797		
Total	1743.996	19			

Table A4.1.6.18: One-way ANOVA table for fresh weight (d 8) of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1953.183	3	651.061	22.545	0.000
Within Groups	462.059	16	28.879		
Total	2415.243	19			

Table A4.1.6.19: One-way ANOVA table for fresh weight (d 8) of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2621.815	3	873.938	9.043	0.001
Within Groups	1546.214	16	96.638		
Total	4168.029	19			

Table A4.1.6.20: One-way ANOVA table for fresh weight (d 8) of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1531.773	3	510.591	5.305	0.010
Within Groups	1539.927	16	96.245		
Total	3071.699	19			

Table A4.1.6.21: One-way ANOVA table for fresh weight (d 8) of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1409.705	3	469.902	18.457	0.000
Within Groups	407.343	16	25.459		
Total	1817.047	19			

Table A4.1.6.22: One-way ANOVA table for fresh weight (d 8) of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2626.290	3	875.430	9.315	0.001
Within Groups	1503.713	16	93.982		
Total	4130.003	19			

Table A4.1.6.23: One-way ANOVA table for fresh weight (d 8) of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	6671.331	3	2223.777	5.843	0.007
Within Groups	6089.607	16	380.600		
Total	12760.939	19			

A4.1.7: Solution usage and fresh weight during vase life

Table A4.1.7.1: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 2).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	466.121	3	155.374	78.220	0.000
Within Groups	31.782	16	1.986		
Total	497.903	19			

Table A4.1.7.2: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 4).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1304.586	3	434.862	45.518	0.000
Within Groups	152.857	16	9.554		
Total	1457.443	19			

Table A4.1.7.3: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 6).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1770.967	3	590.322	87.945	0.000
Within Groups	107.398	16	6.712		
Total	1878.365	19			

Table A4.1.7.4: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 8).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1354.569	3	451.523	24.481	0.000
Within Groups	295.105	16	18.444		
Total	1649.674	19			

Table A4.1.7.5: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 10).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	771.831	3	257.277	19.035	0.000
Within Groups	216.258	16	13.516		
Total	988.088	19			

Table A4.1.7.6: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 12).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	480.874	3	160.291	11.921	0.000
Within Groups	215.135	16	13.446		
Total	696.009	19			

Table A4.1.7.7: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 14).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	153.330	3	51.110	3.694	0.034
Within Groups	221.369	16	13.836		
Total	374.698	19			

Table A4.1.7.8: One-way ANOVA table for fresh weight of 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 16).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	22.719	3	7.573	0.604	0.622
Within Groups	200.460	16	12.529		
Total	223.179	19			

Table A4.1.7.9: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 2).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	514.820	3	171.607	22.315	0.000
Within Groups	123.045	16	7.690		
Total	637.865	19			

Table A4.1.7.10: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 4).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1164.875	3	388.292	31.429	0.000
Within Groups	197.672	16	12.355		
Total	1362.547	19			

Table A4.1.7.11: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 6).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1937.727	3	645.909	39.863	0.000
Within Groups	259.250	16	16.203		
Total	2196.977	19			

Table A4.1.7.12: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 8).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2230.229	3	743.410	27.624	0.000
Within Groups	430.585	16	26.912		
Total	2660.814	19			

Table A4.1.7.13: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 10).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2164.460	3	721.487	23.067	0.000
Within Groups	500.449	16	31.278		
Total	2664.908	19			

Table A4.1.7.14: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 12).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1686.216	3	562.072	17.932	0.000
Within Groups	501.511	16	31.344		
Total	2187.727	19			

Table A4.1.7.15: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 14).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1029.084	3	343.028	10.998	0.000
Within Groups	499.062	16	31.191		
Total	1528.146	19			

Table A4.1.7.16: One-way ANOVA table for fresh weight of 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 16).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	634.321	3	211.440	7.153	0.003
Within Groups	472.939	16	29.559		
Total	1107.261	19			

Table A4.1.7.17: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 1).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.065	3	0.022	2.836	0.071
Within Groups	0.124	16	0.007		
Total	0.190	19			

Table A4.1.7.18: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 3).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.040	3	0.013	0.470	0.707
Within Groups	0.459	16	0.028		
Total	0.500	19			

Table A4.1.7.19: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 5).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.231	3	0.076	10.845	0.000
Within Groups	0.114	16	0.007		
Total	0.344	19			

Table A4.1.7.20: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 7).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.123	3	0.040	1.629	0.222
Within Groups	0.402	16	0.025		
Total	0.525	19			

Table A4.1.7.21: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 9).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.025	3	0.0085	9.030	0.001
Within Groups	0.015	16	0.0009		
Total	0.041	19			

Table A4.1.7.22: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 11).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.035	3	0.011	1.073	0.389
Within Groups	0.179	16	0.011		
Total	0.215	19			

Table A4.1.7.23: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 13).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.056	3	0.0186	62.152	0.000
Within Groups	0.004	16	0.0004		
Total	0.060	19			

Table A4.1.7.24: One-way ANOVA table for solution usage by 'First Red' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 15).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.027	3	0.0092	75.100	0.000
Within Groups	0.004	16	0.0001		
Total	0.029	19			

Table A4.1.7.25: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 1).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.324	3	0.108	4.322	0.021
Within Groups	0.400	16	0.025		
Total	0.724	19			

Table A4.1.7.26: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 3).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.387	3	0.129	34.903	0.000
Within Groups	0.059	16	0.003		
Total	0.447	19			

Table A4.1.7.27: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 5).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.395	3	0.132	24.730	0.000
Within Groups	0.085	16	0.005		
Total	0.480	19			

Table A4.1.7.28: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 7).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.260	3	0.086	21.385	0.000
Within Groups	0.064	16	0.004		
Total	0.325	19			

Table A4.1.7.29: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 9).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.137	3	0.045	12.874	0.000
Within Groups	0.056	16	0.003		
Total	0.194	19			

Table A4.1.7.30: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 11).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.498	3	0.166	16.818	0.000
Within Groups	0.158	16	0.009		
Total	0.656	19			

Table A4.1.7.31: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 13).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.488	3	0.163	28.478	0.000
Within Groups	0.091	16	0.005		
Total	0.580	19			

Table A4.1.7.32: One-way ANOVA table for solution usage by 'Akito' roses stored at 1, 5, and 10°C and for control (non-stored) roses (day 15).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.208	3	0.069	29.760	0.000
Within Groups	0.037	16	0.002		
Total	0.245	19			

A4.1.8: Final pH

Table A4.1.8.1: ANOVA table for solution pH of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	1.096	3	0.365	6.672	0.001
Storage treatment (B)	3.162	3	1.054	19.243	0.000
AxB	0.497	9	0.055	1.007	0.444
Error	3.505	64	0.054		
Total	8.260	79			

Table A4.1.8.2: Solution pH means of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5	6
spring5 [b]	20	3.4324					
spring10	20	3.4878	3.4878				
spring1	20	3.5104	3.5104				
summer5	20	3.5180	3.5180				
summer1	20	3.6480	3.6480	3.6480			
winter10	20	3.6900	3.6900	3.6900	3.6900		
autumn1	20	3.7020	3.7020	3.7020	3.7020		
summer10	20	3.7140	3.7140	3.7140	3.7140		
autumn5	20	3.7290	3.7290	3.7290	3.7290	3.7290	
winter5	20	3.7400	3.7400	3.7400	3.7400	3.7400	
winter1	20		3.8200	3.8200	3.8200	3.8200	
autumn10	20		3.8380	3.8380	3.8380	3.8380	
summerC	20			3.9320	3.9320	3.9320	
springC	20				4.0160	4.0160	
autumnC	20					4.0640	
winterC	20						4.4140
Sig.		0.085	0.050	0.109	0.065	0.053	1.000

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.8.3: ANOVA table for solution pH of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.092	3	0.030	1.658	0.185
Storage treatment (B)	2.467	3	0.822	44.331	0.000
AxB	0.177	9	0.019	1.060	0.404
Error	1.187	64	0.018		
Total	3.923	79			

Table A4.1.8.4: Solution pH means of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
spring5 [b]	20	3.4324			
spring10	20	3.4878	3.4878		
spring1	20	3.5104	3.5104		
summer5	20	3.5180	3.5180	3.5180	
autumn5	20	3.5210	3.5210	3.5210	
winter5	20	3.5320	3.5320	3.5320	
winter1	20	3.5600	3.5600	3.5600	
autumn10	20	3.5810	3.5810	3.5810	
autumn1	20	3.6240	3.6240	3.6240	
winter10	20	3.6240	3.6240	3.6240	
summer1	20		3.6480	3.6480	
summer10	20			3.7140	
winterC	20				3.8940
summerC	20				3.9320
autumnC	20				3.9920
springC	20				4.0160
Sig.		0.065	0.124	0.057	0.203

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.8.5: One-way ANOVA table for solution pH of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.514	3	0.171	2.973	0.063
Within Groups	0.922	16	0.057		
Total	1.435	19			

Table A4.1.8.6: One-way ANOVA table for solution pH of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.853	3	0.618	3.493	0.040
Within Groups	2.829	16	0.177		
Total	4.682	19			

Table A4.1.8.7: One-way ANOVA table for solution pH of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.407	3	0.136	2.109	0.139
Within Groups	1.029	16	0.064		
Total	1.436	19			

Table A4.1.8.8: One-way ANOVA table for solution pH of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.696	3	0.565	5.564	0.008
Within Groups	1.626	16	0.102		
Total	3.322	19			

Table A4.1.8.9: One-way ANOVA table for solution pH of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.106	3	0.369	12.735	0.000
Within Groups	0.463	16	20.028		
Total	1.569	19			

Table A4.1.8.10: One-way ANOVA table for solution pH of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.449	3	0.150	6.186	0.005
Within Groups	0.387	16	0.024		
Total	0.836	19			

Table A4.1.8.11: One-way ANOVA table for solution pH of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.678	3	0.226	33.741	0.000
Within Groups	0.107	16	0.006		
Total	0.785	19			

Table A4.1.8.12: One-way ANOVA table for solution pH of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.411	3	0.137	9.543	0.001
Within Groups	0.230	16	0.014		
Total	0.641	19			

A4.1.9: Solution absorbance and turbidity

Table A4.1.9.1: ANOVA table for solution absorbance of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.01385	3	0.00461	36.263	0.000
Storage treatment (B)	0.00016	3	0.00005	0.435	0.729
AxB	0.00154	9	0.00017	1.344	0.233
Error	0.00814	64	0.01273		
Total	28.700	79			

Table A4.1.9.2: ANOVA table for solution absorbance of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.00846	3	0.00282	31.107	0.000
Storage treatment (B)	0.00011	3	0.00003	0.405	0.750
AxB	0.00029	9	0.00003	0.367	0.947
Error	0.00580	64	0.00009		
Total	0.01469	79			

Table A4.1.9.3: ANOVA table for solution turbidity of 'First Red' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	2.575	3	0.858	3.952	0.012
Storage treatment (B)	2.775	3	0.925	4.259	0.008
AxB	9.450	9	1.050	4.835	0.000
Error	13.900	64	0.217		
Total	28.700	79			

Table A4.1.9.4: ANOVA table for solution turbidity of 'Akito' roses grown during the year and then stored at 1, 5, and 10°C and for control (non-stored) flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	1.409	3	0.470	2.244	0.092
Storage treatment (B)	3.784	3	1.261	6.025	0.001
AxB	1.153	9	0.128	0.612	0.782
Error	13.400	64	0.209		
Total	19.747	79			

Table A4.1.9.5: Solution absorbance means of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5
autumn10 [b]	20	0.0025				
autumn1	20	0.0028				
autumn5	20	0.0051	0.00513			
autumnC	20	0.0167	0.01670	0.01670		
winter10	20	0.0175	0.01753	0.01753		
winterC	20		0.01870	0.01870		
winter1	20		0.01963	0.01963		
winter5	20		0.01967	0.01967		
springC	20			0.02650	0.02650	
spring5	20			0.03153	0.03153	0.03153
summer5	20				0.03553	0.03553
summerC	20				0.03650	0.03650
spring1	20				0.03900	0.03900
spring10	20				0.04027	0.04027
summer10	20					0.04427
summer1	20					0.04519
Sig.		0.063	0.078	0.076	0.095	0.103

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.9.6: Solution turbidity means of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
autumn1 [b]	20	1.0000			
autumn5	20	1.0000			
winterC	20	1.0000			
springC	20	1.0000			
winter1	20	1.1000			
winter5	20	1.1000			
autumn10	20	1.2000			
spring5	20	1.2000			
summer10	20	1.3000			
spring1	20	1.4000	1.4000		
summer5	20	1.4000	1.4000		
winter10	20	1.5000	1.5000		
summer1	20	1.6000	1.6000	1.6000	
spring10	20		2.0000	2.0000	2.0000
autumnC	20			2.2000	2.2000
summerC	20				2.4000
Sig.		0.097	0.073	0.058	0.206

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.9.7: Solution turbidity means of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were control (non-stored flowers) or stored at 1, 5 and 10°C for 10 days. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
autumn1 [b]	20	1.0000		
autumn5	20	1.0000		
winter5	20	1.0000		
winter1	20	1.1000		
summer5	20	1.1000		
autumn10	20	1.2000	1.2000	
spring1	20	1.2000	1.2000	
spring10	20	1.2000	1.2000	
summer1	20	1.2000	1.2000	
winter10	20	1.3000	1.3000	
summer10	20	1.3000	1.3000	
autumnC	20	1.4000	1.4000	1.4000
winterC	20	1.5000	1.5000	1.5000
spring5	20	1.6000	1.6000	1.6000
summerC	20		1.8000	1.8000
springC	20			2.0000
Sig.		0.092	0.085	0.068

[a] autumn, winter, spring, summer = growing seasons during the year. [b] C, 1, 5, 10 = Control, 1, 5, 10°C, respectively.

Table A4.1.9.8: One-way ANOVA table for solution absorbance of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.400	3	1.467	8.092	0.002
Within Groups	2.900	16	0.181		
Total	7.300	19			

Table A4.1.9.9: One-way ANOVA table for solution absorbance of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.737	3	0.246	2.810	0.073
Within Groups	1.400	16	0.087		
Total	2.137	19			

Table A4.1.9.10: One-way ANOVA table for solution absorbance of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.800	3	0.933	1.867	0.176
Within Groups	8.000	16	0.500		
Total	10.800	19			

Table A4.1.9.11: One-way ANOVA table for solution absorbance of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00067	3	0.00022	2.907	0.067
Within Groups	0.00124	16	0.00007		
Total	0.00191	19			

Table A4.1.9.12: One-way ANOVA table for solution absorbance of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00023	3	0.00007	0.577	0.639
Within Groups	0.00217	16	0.00013		
Total	0.00240	19			

Table A4.1.9.13: One-way ANOVA table for solution absorbance of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00023	3	0.00007	0.577	0.639
Within Groups	0.00217	16	0.00013		
Total	0.00240	19			

Table A4.1.9.14: One-way ANOVA table for solution absorbance of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00006	3	0.00002	0.157	0.924
Within Groups	0.00209	16	0.00013		
Total	0.00215	19			

Table A4.1.9.15: One-way ANOVA table for solution absorbance of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00003	3	0.00001	0.182	0.907
Within Groups	0.00111	16	0.00006		
Total	0.00115	19			

Table A4.1.9.16: One-way ANOVA table for solution turbidity of 'First Red' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.737	3	0.246	2.810	0.073
Within Groups	1.400	16	0.087		
Total	2.137	19			

Table A4.1.9.17: One-way ANOVA table for solution turbidity of 'First Red' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.800	3	0.933	1.867	0.176
Within Groups	8.000	16	0.500		
Total	10.800	19			

Table A4.1.9.18: One-way ANOVA table for solution turbidity of 'First Red' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3.737	3	1.246	8.306	0.001
Within Groups	2.400	16	0.150		
Total	6.137	19			

Table A4.1.9.19: One-way ANOVA table for solution turbidity of 'First Red' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.400	3	1.467	8.092	0.002
Within Groups	2.900	16	0.181		
Total	7.300	19			

Table A4.1.9.20: One-way ANOVA table for solution turbidity of 'Akito' roses grown in winter and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.737	3	0.246	1.967	0.160
Within Groups	2.000	16	0.125		
Total	2.738	19			

Table A4.1.9.21: One-way ANOVA table for solution turbidity of 'Akito' roses grown in spring and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2.200	3	0.733	1.333	0.299
Within Groups	8.800	16	0.550		
Total	11.000	19			

Table A4.1.9.22: One-way ANOVA table for solution turbidity of 'Akito' roses grown in summer and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.450	3	0.483	4.833	0.014
Within Groups	1.600	16	0.100		
Total	3.050	19			

Table A4.1.9.23: One-way ANOVA table for solution turbidity of 'Akito' roses grown in autumn and then stored at 1, 5, and 10°C and for control (non-stored) roses.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.550	3	0.183	2.933	0.065
Within Groups	1.000	16	0.062		
Total	1.550	19			

APPENDIX 4.2: WATER LOSS AND DRY WEIGHT OF DETACHED LEAVES

A4.2.1: Water loss

Table A4.2.1.1: ANOVA table for water loss of detached leaves from rose stems at 1, 5, and 10°C and 75, 85 and 95% RH throughout the year. Data are means of water loss measured 12, 24 and 48 h after detachment.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	55499.033	3	18499.678	252.513	0.000
Temperature (B)	2949.998	2	1474.999	20.133	0.000
Relative Humidity (C)	6429.828	2	3214.914	43.882	0.000
Cultivar (D)	5721.684	1	5721.684	78.099	0.000
A x B	400.393	6	66.732	0.911	0.487
A x C	1213.534	6	202.256	2.761	0.013
A X D	4260.168	3	1420.056	19.383	0.000
B x C	277.160	4	69.290	0.946	0.438
B x D	372.014	2	186.007	2.539	0.081
C x D	316.874	2	158.437	2.163	0.117
A x B x C	957.038	12	79.753	1.089	0.369
A x B x D	1217.934	6	202.989	2.771	0.012
A x C x D	327.723	6	54.621	0.746	0.613
B x C x D	102.608	4	25.652	0.350	0.844
A x B x C x D	888.264	12	74.022	1.010	0.439
Error	21099.495	288	73.262		
Total	102033.748	359			

Table A4.2.1.2: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	476.339	2	238.170	4.888	0.028
Within Groups	584.730	12	48.728		
Total	1061.069	14			

Table A4.2.1.3: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	572.309	2	286.155	4.400	0.037
Within Groups	780.475	12	65.040		
Total	1352.784	14			

Table A4.2.1.4: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	632.222	2	316.111	6.046	0.015
Within Groups	627.393	12	52.283		
Total	1259.615	14			

Table A4.2.1.5: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	170.317	2	85.158	0.193	0.827
Within Groups	5303.276	12	441.940		
Total	5473.593	14			

Table A4.2.1.6: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	161.514	2	80.757	4.301	0.039
Within Groups	225.334	12	18.778		
Total	386.849	14			

Table A4.2.1.7: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	182.018	2	91.009	6.740	0.011
Within Groups	162.038	12	13.503		
Total	344.057	14			

Table A4.2.1.8: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	132.001	2	66.001	2.100	0.165
Within Groups	377.063	12	31.422		
Total	509.064	14			

Table A4.2.1.9: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	134.489	2	67.244	0.277	0.763
Within Groups	2911.211	12	242.601		
Total	3045.700	14			

Table A4.2.1.10: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	225.974	2	112.987	5.078	0.025
Within Groups	267.004	12	22.250		
Total	492.978	14			

Table A4.2.1.11: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	315.162	2	157.581	15.849	0.000
Within Groups	119.310	12	9.943		
Total	434.472	14			

Table A4.2.1.12: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	95.386	2	47.693	0.500	0.619
Within Groups	1145.441	12	95.453		
Total	1240.827	14			

Table A4.2.1.13: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	509.946	2	254.973	1.009	0.393
Within Groups	3031.485	12	252.624		
Total	3541.431	14			

Table A4.2.1.14: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	105.078	2	52.539	0.710	0.511
Within Groups	888.181	12	74.015		
Total	993.259	14			

Table A4.2.1.15: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	456.534	2	228.267	3.527	0.062
Within Groups	776.545	12	64.712		
Total	1233.079	14			

Table A4.2.1.16: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	666.525	2	333.263	8.807	0.004
Within Groups	454.069	12	37.839		
Total	1120.594	14			

Table A4.2.1.17: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	523.320	2	261.660	6.198	0.014
Within Groups	506.611	12	42.218		
Total	1029.932	14			

Table A4.2.1.18: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	87.885	2	43.942	1.299	0.308
Within Groups	405.864	12	33.822		
Total	493.749	14			

Table A4.2.1.19: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	234.042	2	117.021	2.857	0.097
Within Groups	491.596	12	40.966		
Total	725.638	14			

Table A4.2.1.20: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	196.068	2	98.034	1.329	0.301
Within Groups	885.180	12	73.765		
Total	1081.248	14			

Table A4.2.1.21: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	852.724	2	426.362	6.881	0.010
Within Groups	743.551	12	61.963		
Total	1596.275	14			

Table A4.2.1.22: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	133.017	2	66.509	1.734	0.218
Within Groups	460.337	12	38.361		
Total	593.355	14			

Table A4.2.1.23: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	104.183	2	52.091	5.564	0.020
Within Groups	112.348	12	9.362		
Total	216.530	14			

Table A4.2.1.24: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	218.316	2	109.158	3.282	0.073
Within Groups	399.161	12	33.263		
Total	617.477	14			

Table A4.2.1.25: One-way ANOVA table for water loss of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	342.891	2	171.446	3.163	0.079
Within Groups	650.451	12	54.204		
Total	993.342	14			

A4.2.2: Dry weight

Table A4.2.2.1: ANOVA table for dry weight of detached leaves from rose stems at 1, 5, and 10°C and 75, 85 and 95% RH throughout the year. Data are means of water loss measured 12, 24 and 48 h after detachment.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Growing seasons (A)	0.334	3	0.111	14.981	0.000
Temperature (B)	0.0092	2	0.0046	0.624	0.537
Relative Humidity (C)	0.107	2	0.0534	7.190	0.001
Cultivar (D)	4.773	1	4.773	642.281	0.000
A x B	0.0367	6	0.0061	0.825	0.551
A x C	0.0843	6	0.0140	1.893	0.082
A X D	0.134	3	0.0445	5.995	0.001
B x C	0.136	4	0.0340	4.579	0.001
B x D	0.0833	2	0.0416	5.610	0.004
C x D	0.0119	2	0.059	0.807	0.447
A x B x C	0.107	12	0.0089	1.205	0.279
A x B x D	0.085	6	0.014	1.913	0.079
A x C x D	0.0544	6	0.0090	1.222	0.295
B x C x D	0.0434	4	0.0108	1.461	0.214
A x B x C x D	0.0826	12	0.0068	0.927	0.520
Error	2.140	288	0.0074		
Total	8.222	359			

Table A4.2.2.2: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.152	2	0.075	4.060	0.045
Within Groups	0.224	12	0.018		
Total	0.376	14			

Table A4.2.2.3: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.152	2	0.075	6.198	0.014
Within Groups	0.147	12	0.012		
Total	0.299	14			

Table A4.2.2.4: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.008	2	0.004	0.457	0.644
Within Groups	0.114	12	0.009		
Total	0.123	14			

Table A4.2.2.5: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'First Red' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.008	2	0.004	.0502	0.618
Within Groups	0.104	12	0.008		
Total	0.113	14			

Table A4.2.2.6: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0009	2	0.0004	0.057	0.945
Within Groups	0.0982	12	0.0081		
Total	0.0991	14			

Table A4.2.2.7: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0009	2	0.0004	0.070	0.933
Within Groups	0.0794	12	0.0066		
Total	0.0803	14			

Table A4.2.2.8: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0542	2	0.0271	2.259	0.147
Within Groups	0.144	12	0.0120		
Total	0.198	14			

Table A4.2.2.9: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'First Red' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.125	2	0.0624	6.079	0.015
Within Groups	0.123	12	0.0102		
Total	0.248	14			

Table A4.2.2.10: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0012	2	0.0004	0.062	0.940
Within Groups	0.118	12	0.0098		
Total	0.119	14			

Table A4.2.2.11: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0022	2	0.0011	0.142	0.869
Within Groups	0.0966	12	0.0080		
Total	0.0989	14			

Table A4.2.2.12: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0021	2	0.0010	0.134	0.876
Within Groups	0.0975	12	0.0081		
Total	0.0996	14			

Table A4.2.2.13: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'First Red' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0021	2	0.0010	0.150	0.862
Within Groups	0.0869	12	0.0072		
Total	0.0890	14			

Table A4.2.2.14: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.002	2	0.001	0.194	0.826
Within Groups	0.084	12	0.007		
Total	0.087	14			

Table A4.2.2.15: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.002	2	0.001	0.372	0.697
Within Groups	0.044	12	0.003		
Total	0.047	14			

Table A4.2.2.16: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.010	2	0.005	2.523	0.122
Within Groups	0.025	12	0.002		
Total	0.036	14			

Table A4.2.2.17: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 75% RH of 'Akito' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.014	2	0.007	1.715	0.221
Within Groups	0.049	12	0.004		
Total	0.063	14			

Table A4.2.2.18: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.007	2	0.003	0.653	0.538
Within Groups	0.066	12	0.005		
Total	0.074	14			

Table A4.2.2.19: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.007	2	0.003	1.199	0.335
Within Groups	0.036	12	0.003		
Total	0.043	14			

Table A4.2.2.20: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.011	2	0.005	2.137	0.161
Within Groups	0.032	12	0.002		
Total	0.043	14			

Table A4.2.2.21: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 85% RH of 'Akito' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.007	2	0.003	0.562	0.584
Within Groups	0.084	12	0.007		
Total	0.092	14			

Table A4.2.2.22: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.018	2	0.009	1.341	0.298
Within Groups	0.084	12	0.007		
Total	0.103	14			

Table A4.2.2.23: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in spring.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.018	2	0.009	3.288	0.073
Within Groups	0.034	12	0.002		
Total	0.053	14			

Table A4.2.2.24: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in summer.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.023	2	0.011	5.383	0.021
Within Groups	0.025	12	0.002		
Total	0.048	14			

Table A4.2.2.25: One-way ANOVA table for dry weight of detached leaves at 1, 5, and 10°C and 95% RH of 'Akito' roses grown in autumn.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.024	2	0.012	2.425	0.130
Within Groups	0.059	12	0.004		
Total	0.083	14			

APPENDIX 5

EFFECT OF DIFFERENT ABSCISIC ACID TREATMENTS TO IMPROVE VASE LIFE OF CUT 'FIRST RED' AND 'AKITO' ROSES STORED AT LOW TEMPERATURE

APPENDIX 5.1: EFFECTS OF STORAGE TEMPERATURE AND ABA TREATMENTS ON VASE LIFE

A5.1.1: Flower life

Table A5.1.1.1: ANOVA table for flower life of 'First Red' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	56.000	2	28.000	13.500	0.000
ABA treatment (B)	0.222	2	0.111	0.054	0.948
AxB	23.111	4	5.778	2.786	0.058
Error	37.333	18	2.074		
Total	116.667	26			

Table A5.1.1.2: Flower life means of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
5S [b]	12	6.0000		
5P	12	6.0000		
1C	12	6.6667		
1S	12	7.3333	7.3333	
1P	12	8.6667	8.6667	8.6667
5C	12	8.6667	8.6667	8.6667
NOC	12		9.6667	9.6667
NOP	12		10.0000	10.0000
NOS	12			11.0000
Sig.		0.058	0.054	0.089

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.1.1.3: ANOVA table for flower life of 'Akito' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	3.630	2	1.815	0.302	0.743
ABA treatment (B)	30.519	2	15.259	2.543	0.106
AxB	50.815	4	12.704	2.117	0.121
Error	108.000	18	6.000		
Total	192.963	26			

A5.1.2: Foliage life

Table A5.1.2.1: ANOVA table for foliage life of 'First Red' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	137.556	2	68.778	12.633	0.000
ABA treatment (B)	14.889	2	7.444	1.367	0.280
AxB	42.222	4	10.556	1.939	0.148
Error	98.000	18	5.444		
Total	292.667	26			

Table A5.1.2.2: Foliage life means of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
5C [b]	12	5.6667		
5P	12	7.0000	7.0000	
1P	12	8.0000	8.0000	
1S	12	9.6667	9.6667	9.6667
5S	12		10.6667	10.6667
1C	12		11.3333	11.3333
NOC	12			13.0000
NOS	12			13.3333
NOP	12			13.3333
Sig.		0.068	0.054	0.103

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.1.2.3: ANOVA table for foliage life of 'Akito' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	110.519	2	55.259	4.722	0.022
ABA treatment (B)	58.074	2	29.037	2.481	0.112
AxB	237.926	4	59.481	5.082	0.006
Error	210.667	18	11.704		
Total	617.185	26			

Table A5.1.2.4: Foliage life means of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
5C [b]	12	5.0000			
1P	12	7.0000	7.0000		
1C	12	7.6667	7.6667		
1S	12	7.6667	7.6667		
NOS	12	9.0000	9.0000	9.0000	
5S	12	11.0000	11.0000	11.0000	
NOP	12		12.3333	12.3333	12.3333
NOC	12			15.0000	15.0000
5P	12				17.6667
Sig.		0.071	0.106	0.062	0.086

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

APPENDIX 5.2: EFFECTS OF STORAGE TEMPERATURE AND ABA TREATMENTS ON F_v/F_m

A5.2.1: F_v/F_m on day 0

Table A5.2.1.1: ANOVA table for leaf F_v/F_m on d 0 of 'First Red' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	0.081	2	0.040	29.953	0.000
ABA treatment (B)	0.011	2	0.081	4.196	0.032
AxB	0.014	4	0.011	2.707	0.063
Error	0.024	18	0.014		
Total	0.132	26			

Table A5.2.1.2: F_v/F_m means of 'First Red' roses on d 0 separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4	5
1C [b]	12	0.64933				
1P	12	0.68867	0.68867			
5P	12		0.73300	0.73300		
1S	12		0.75233	0.75233	0.75233	
5C	12			0.79700	0.79700	0.79700
5S	12				0.80567	0.80567
NOP	12					0.82200
NOS	12					0.83133
NOC	12					0.83733
Sig.		0.208	0.059	0.058	0.110	0.243

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.2.1.3: ANOVA table for leaf F_v/F_m on d 0 of 'Akito' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	0.0018	2	0.0009	2.070	0.155
ABA treatment (B)	0.0013	2	0.0006	1.548	0.240
AxB	0.0011	4	0.0002	0.624	0.652
Error	0.0080	18	0.0004		
Total	0.0124	26			

A5.2.2: F_v/F_m during vase life

Table A5.2.2.1: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0587	2	0.0293	33.000	0.001
Within Groups	0.0053	6	0.0009		
Total	0.0641	8			

Table A5.2.2.2: One-way ANOVA table for F_v/F_m on d 4 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.089	2	0.044	4.004	0.079
Within Groups	0.066	6	0.011		
Total	0.156	8			

Table A5.2.2.3: One-way ANOVA table for F_v/F_m on d 8 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.129	2	0.064	5.606	0.042
Within Groups	0.068	6	0.011		
Total	0.197	8			

Table A5.2.2.4: One-way ANOVA table for F_v/F_m on d 12 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.085	2	0.042	4.214	0.072
Within Groups	0.060	6	0.010		
Total	0.146	8			

Table A5.2.2.5: One-way ANOVA table for F_v/F_m on d 16 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.081	2	0.040	3.796	0.086
Within Groups	0.064	6	0.010		
Total	0.146	8			

Table A5.2.2.6: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.009	2	0.004	2.081	0.206
Within Groups	0.014	6	0.002		
Total	0.023	8			

Table A5.2.2.7: One-way ANOVA table for F_v/F_m on d 4 of 'First Red' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.014	2	0.007	1.875	0.233
Within Groups	0.023	6	0.003		
Total	0.038	8			

Table A5.2.2.8: One-way ANOVA table for F_v/F_m on d 8 of 'First Red' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.006	2	0.003	1.331	0.332
Within Groups	0.015	6	0.002		
Total	0.021	8			

Table A5.2.2.9: One-way ANOVA table for F_v/F_m on d 12 of 'First Red' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.004	2	0.002	0.942	0.441
Within Groups	0.014	6	0.002		
Total	0.018	8			

Table A5.2.2.10: One-way ANOVA table for F_v/F_m on d 16 of 'First Red' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.003	2	0.001	0.765	0.506
Within Groups	0.013	6	0.002		
Total	0.017	8			

Table A5.2.2.11: One-way ANOVA table for F_v/F_m on d 0 of 'First Red' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0276	2	0.0138	16.220	0.004
Within Groups	0.0051	6	0.0008		
Total	0.0327	8			

Table A5.2.2.12: One-way ANOVA table for F_v/F_m on d 4 of 'First Red' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0202	2	0.0101	11.944	0.008
Within Groups	0.0050	6	0.0008		
Total	0.0253	8			

Table A5.2.2.13: One-way ANOVA table for F_v/F_m on d 8 of 'First Red' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.004	2	0.002	0.251	0.786
Within Groups	0.054	6	0.009		
Total	0.059	8			

Table A5.2.2.14: One-way ANOVA table for F_v/F_m on d 12 of 'First Red' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.010	2	0.005	0.403	0.685
Within Groups	0.077	6	0.012		
Total	0.087	8			

Table A5.2.2.15: One-way ANOVA table for F_v/F_m on d 16 of 'First Red' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.009	2	0.004	0.405	0.684
Within Groups	0.073	6	0.012		
Total	0.083	8			

Table A5.2.2.16: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00030	2	0.00015	15.621	0.004
Within Groups	0.00005	6	0.000009		
Total	0.00036	8			

Table A5.2.2.17: One-way ANOVA table for F_v/F_m on d 4 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00020	2	0.00010	2.321	0.179
Within Groups	0.00025	6	0.00004		
Total	0.00046	8			

Table A5.2.2.18: One-way ANOVA table for F_v/F_m on d 8 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00032	2	0.00016	1.125	0.385
Within Groups	0.00085	6	0.00014		
Total	0.00117	8			

Table A5.2.2.19: One-way ANOVA table for F_v/F_m on d 12 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.020	2	0.010	0.650	0.555
Within Groups	0.096	6	0.016		
Total	0.117	8			

Table A5.2.2.20: One-way ANOVA table for F_v/F_m on d 16 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA before storage (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.025	2	0.012	0.807	0.489
Within Groups	0.093	6	0.015		
Total	0.118	8			

Table A5.2.2.21: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0008	2	0.0004	1.293	0.341
Within Groups	0.0020	6	0.0003		
Total	0.0028	8			

Table A5.2.2.22: One-way ANOVA table for F_v/F_m on d 4 of 'Akito' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00013	2	0.00006	4.203	0.072
Within Groups	0.00009	6	0.00001		
Total	0.00022	8			

Table A5.2.2.23: One-way ANOVA table for F_v/F_m on d 8 of 'Akito' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0005	2	0.0002	2.436	0.168
Within Groups	0.0006	6	0.0001		
Total	0.0011	8			

Table A5.2.2.24: One-way ANOVA table for F_v/F_m on d 12 of 'Akito' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.024	2	0.012	0.986	0.426
Within Groups	0.075	6	0.012		
Total	0.100	8			

Table A5.2.2.25: One-way ANOVA table for F_v/F_m on d 16 of 'Akito' roses sprayed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.025	2	0.012	0.515	0.622
Within Groups	0.148	6	0.024		
Total					

Table A5.2.2.26: One-way ANOVA table for F_v/F_m on d 0 of 'Akito' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0018	2	0.0009	0.911	0.451
Within Groups	0.0060	6	0.0010		
Total	0.0078	8			

Table A5.2.2.27: One-way ANOVA table for F_v/F_m on d 4 of 'Akito' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.016	2	0.008	0.820	0.484
Within Groups	0.062	6	0.010		
Total	0.079	8			

Table A5.2.2.28: One-way ANOVA table for F_v/F_m on d 8 of 'Akito' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.034	2	0.017	1.291	0.342
Within Groups	0.080	6	0.013		
Total	0.116	8			

Table A5.2.2.29: One-way ANOVA table for F_v/F_m on d 12 of 'Akito' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.022	2	0.011	1.566	1.595
Within Groups	0.118	6	0.019		
Total	0.140	8			

Table A5.2.2.30: One-way ANOVA table for F_v/F_m on d 16 of 'Akito' roses pulsed with 10^{-5} M ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.024	2	0.012	0.765	0.506
Within Groups	0.096	6	0.016		
Total	0.121	8			

APPENDIX 5.3: EFFECTS OF STORAGE TEMPERATURE AND ABA TREATMENTS ON FLOWER CONDITION

A5.3.1 Bent neck

Table A5.3.1.1: ANOVA table for bent neck of 'First Red' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	740.741	2	370.370	0.571	0.575
ABA treatment (B)	185.185	2	92.593	0.143	0.868
AxB	1481.481	4	370.370	0.571	0.687
Error	11666.667	18	648.148		
Total	14074.074	26			

Table A5.3.1.2: ANOVA table for bent neck of 'Akito' roses treated with different ABA solutions and then stored at 1 and 5°C and for non-stored flowers.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	2962.963	2	1481.481	0.500	0.615
ABA treatment (B)	740.741	2	370.370	0.125	0.883
AxB	5925.926	4	1481.481	0.500	0.736
Error	53333.333	18	2962.963		
Total	62962.963	26			

A5.3.2 Corolla diameter during vase life

Table A5.3.2.1: One-way ANOVA table for corolla diameter on d 0 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	550.389	2	275.194	2.752	0.142
Within Groups	600.000	6	100.000		
Total	1150.389	8			

Table A5.3.2.2: One-way ANOVA table for corolla diameter on d 4 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	833.625	2	416.813	4.519	0.064
Within Groups	553.375	6	92.229		
Total	1387.000	8			

Table A5.3.2.3: One-way ANOVA table for corolla diameter on d 8 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	638.097	2	319.049	3.285	0.109
Within Groups	582.708	6	97.118		
Total	1220.806	8			

Table A5.3.2.4: One-way ANOVA table for corolla diameter on d 12 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	235.292	2	117.646	2.355	0.176
Within Groups	299.708	6	49.951		
Total	535.000	8			

Table A5.3.2.5: One-way ANOVA table for corolla diameter on d 16 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	310.167	2	155.083	3.675	0.091
Within Groups	253.208	6	42.201		
Total	563.375	8			

Table A5.3.2.6: One-way ANOVA table for corolla diameter on d 0 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	260.722	2	130.361	5.063	0.052
Within Groups	154.500	6	25.750		
Total	415.222	8			

Table A5.3.2.7: One-way ANOVA table for corolla diameter on d 4 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	26.347	2	13.174	0.097	0.909
Within Groups	813.333	6	135.556		
Total	839.681	8			

Table A5.3.2.8: One-way ANOVA table for corolla diameter on d 8 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	9.125	2	4.563	0.028	0.972
Within Groups	969.000	6	161.500		
Total	978.125	8			

Table A5.3.2.9: One-way ANOVA table for corolla diameter on d 12 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	85.847	2	42.924	1.221	0.359
Within Groups	210.958	6	35.160		
Total					

Table A5.3.2.10: One-way ANOVA table for corolla diameter on d 16 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	20.667	2	10.333	0.119	0.890
Within Groups	520.333	6	86.722		
Total	541.000	8			

Table A5.3.2.11: One-way ANOVA table for corolla diameter on d 0 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	25.389	2	12.694	0.211	0.816
Within Groups	361.833	6	60.306		
Total	387.222	8			

Table A5.3.2.12: One-way ANOVA table for corolla diameter on d 4 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	423.014	2	211.507	0.869	0.466
Within Groups	1459.917	6	243.319		
Total	1882.931	8			

Table A5.3.2.13: One-way ANOVA table for corolla diameter on d 8 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	614.847	2	307.424	1.320	0.335
Within Groups	1397.833	6	232.972		
Total	2012.681	8			

Table A5.3.2.14: One-way ANOVA table for corolla diameter on d 12 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	678.514	2	339.257	5.538	0.043
Within Groups	367.542	6	61.257		
Total	1046.056	8			

Table A5.3.2.15: One-way ANOVA table for corolla diameter on d 16 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	226.625	2	113.313	5.317	0.047
Within Groups	127.875	6	21.313		
Total	354.500	8			

Table A5.3.2.16: One-way ANOVA table for corolla diameter on d 0 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	299.556	2	149.778	3.421	0.102
Within Groups	262.667	6	43.778		
Total	562.222	8			

Table A5.3.2.17: One-way ANOVA table for corolla diameter on d 4 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	208.222	2	104.111	0.298	0.752
Within Groups	2094.000	6	349.000		
Total					

Table A5.3.2.18: One-way ANOVA table for corolla diameter on d 8 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	376.222	2	188.111	0.319	0.739
Within Groups	3543.333	6	590.556		
Total	3919.556	8			

Table A5.3.2.19: One-way ANOVA table for corolla diameter on d 12 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2020.847	2	1010.424	2.360	0.175
Within Groups	2568.792	6	428.132		
Total	4589.639	8			

Table A5.3.2.20: One-way ANOVA table for corolla diameter on d 16 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2190.889	2	1095.444	2.385	0.173
Within Groups	2755.333	6	459.222		
Total					

Table A5.3.2.21: One-way ANOVA table for corolla diameter on d 0 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	110.222	2	55.111	0.703	0.532
Within Groups	470.667	6	78.444		
Total	580.889	8			

Table A5.3.2.22: One-way ANOVA table for corolla diameter on d 4 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	16.222	2	8.111	0.018	0.982
Within Groups	2636.000	6	439.333		
Total	2652.222	8			

Table A5.3.2.23: One-way ANOVA table for corolla diameter on d 8 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	17.556	2	8.778	0.014	0.986
Within Groups	3828.000	6	638.000		
Total	3845.556	8			

Table A5.3.2.24: One-way ANOVA table for corolla diameter on d 12 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	460.792	2	230.396	0.318	0.739
Within Groups	4353.458	6	725.576		
Total	4814.250	8			

Table A5.3.2.25: One-way ANOVA table for corolla diameter on d 16 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1536.889	2	768.444	1.326	0.333
Within Groups	3476.667	6	579.444		
Total	5013.556	8			

Table A5.3.2.26: One-way ANOVA table for corolla diameter on d 0 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	88.667	2	44.333	0.476	0.643
Within Groups	559.333	6	93.222		
Total	648.000	8			

Table A5.3.2.27: One-way ANOVA table for corolla diameter on d 4 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	368.222	2	184.111	0.278	0.767
Within Groups	3975.333	6	662.556		
Total	4343.556	8			

Table A5.3.2.28: One-way ANOVA table for corolla diameter on d 8 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	648.667	2	324.333	0.632	0.563
Within Groups	3077.333	6	512.889		
Total	3726.000	8			

Table A5.3.2.29: One-way ANOVA table for corolla diameter on d 12 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	447.056	2	223.528	0.529	0.614
Within Groups	2533.000	6	422.167		
Total	2980.056	8			

Table A5.3.2.30: One-way ANOVA table for corolla diameter on d 16 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1354.667	2	677.333	1.244	0.353
Within Groups	3267.333	6	544.556		
Total	4622.000	8			

APPENDIX 5.4: EFFECTS OF STORAGE TEMPERATURE AND ABA TREATMENTS ON FRESH WEIGHT AND SOLUTION USAGE

A5.4.1: Fresh weight during vase life

Table A5.4.1.1: One-way ANOVA table for fresh weight on d 2 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	226.268	2	113.134	44.267	0.000
Within Groups	15.334	6	2.556		
Total	241.602	8			

Table A5.4.1.2: One-way ANOVA table for fresh weight on d 4 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	360.035	2	180.017	28.735	0.001
Within Groups	37.588	6	6.265		
Total	397.623	8			

Table A5.4.1.3: One-way ANOVA table for fresh weight on d 6 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	470.823	2	235.412	36.633	0.000
Within Groups	38.557	6	6.426		
Total					

Table A5.4.1.4: One-way ANOVA table for fresh weight on d 8 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	494.611	2	247.305	30.894	0.001
Within Groups	48.030	6	8.005		
Total	542.641	8			

Table A5.4.1.5: One-way ANOVA table for fresh weight on d 10 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	690.768	2	345.384	31.518	0.001
Within Groups	65.751	6	10.958		
Total	756.518	8			

Table A5.4.1.6: One-way ANOVA table for fresh weight on d 12 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	397.314	2	198.657	18.187	0.003
Within Groups	65.539	6	10.923		
Total	462.853	8			

Table A5.4.1.7: One-way ANOVA table for fresh weight on d 14 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	235.848	2	117.924	9.816	0.013
Within Groups	72.084	6	12.014		
Total	307.931	8			

Table A5.4.1.8: One-way ANOVA table for fresh weight on d 16 of 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	275.298	2	137.649	9.431	0.014
Within Groups	87.576	6	14.596		
Total	362.875	8			

Table A5.4.1.9: One-way ANOVA table for fresh weight on d 2 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	179.029	2	89.515	62.831	0.000
Within Groups	8.548	6	1.425		
Total	187.577	8			

Table A5.4.1.10: One-way ANOVA table for fresh weight on d 4 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	183.744	2	91.872	19.688	0.002
Within Groups	27.998	6	4.666		
Total	211.742	8			

Table A5.4.1.11: One-way ANOVA table for fresh weight on d 6 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	159.969	2	79.984	4.814	0.057
Within Groups	99.690	6	16.615		
Total	259.659	8			

Table A5.4.1.12: One-way ANOVA table for fresh weight on d 8 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	190.557	2	95.279	8.000	0.020
Within Groups	71.460	6	11.910		
Total	262.017	8			

Table A5.4.1.13: One-way ANOVA table for fresh weight on d 10 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	386.997	2	193.499	14.354	0.005
Within Groups	80.882	6	13.480		
Total	467.879	8			

Table A5.4.1.14: One-way ANOVA table for fresh weight on d 12 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	152.665	2	76.332	5.886	0.038
Within Groups	77.815	6	12.969		
Total	230.480	8			

Table A5.4.1.15: One-way ANOVA table for fresh weight on d 14 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	103.420	2	51.710	3.633	0.093
Within Groups	85.398	6	14.233		
Total	188.818	8			

Table A5.4.1.16: One-way ANOVA table for fresh weight on d 16 of 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	338.293	2	169.146	13.712	0.006
Within Groups	74.012	6	12.335		
Total	412.305	8			

Table A5.4.1.17: One-way ANOVA table for fresh weight on d 2 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	229.178	2	114.589	17.056	0.003
Within Groups	40.312	6	6.719		
Total	269.489	8			

Table A5.4.1.18: One-way ANOVA table for fresh weight on d 4 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	483.432	2	241.716	39.156	0.000
Within Groups	37.038	6	6.173		
Total	520.471	8			

Table A5.4.1.19: One-way ANOVA table for fresh weight on d 6 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	717.566	2	358.783	27.868	0.001
Within Groups	77.246	6	12.874		
Total	794.812	8			

Table A5.4.1.20: One-way ANOVA table for fresh weight on d 8 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	482.894	2	241.447	28.447	0.001
Within Groups	50.927	6	8.488		
Total	533.821	8			

Table A5.4.1.21: One-way ANOVA table for fresh weight on d 10 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	645.253	2	322.626	13.907	0.006
Within Groups	139.194	6	23.199		
Total	784.447	8			

Table A5.4.1.22: One-way ANOVA table for fresh weight on d 12 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	453.303	2	226.652	5.090	0.051
Within Groups	267.147	6	44.524		
Total	720.450	8			

Table A5.4.1.23: One-way ANOVA table for fresh weight on d 14 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	210.422	2	105.211	2.124	0.201
Within Groups	297.175	6	49.529		
Total	507.597	8			

Table A5.4.1.24: One-way ANOVA table for fresh weight on d 16 of 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	408.917	2	204.459	4.169	0.073
Within Groups	294.236	6	49.039		
Total	703.153	8			

Table A5.4.1.25: One-way ANOVA table for fresh weight on d 2 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	100.290	2	50.145	9.740	0.013
Within Groups	30.891	6	5.149		
Total	131.182	8			

Table A5.4.1.26: One-way ANOVA table for fresh weight on d 4 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	158.168	2	79.084	5.805	0.040
Within Groups	81.743	6	13.624		
Total	239.911	8			

Table A5.4.1.27: One-way ANOVA table for fresh weight on d 6 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	75.706	2	37.853	1.016	0.417
Within Groups	223.623	6	37.271		
Total	299.329	8			

Table A5.4.1.28: One-way ANOVA table for fresh weight on d 8 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	99.612	2	49.806	0.430	0.669
Within Groups	694.552	6	115.759		
Total	794.164	8			

Table A5.4.1.29: One-way ANOVA table for fresh weight on d 10 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	155.207	2	77.603	0.503	0.628
Within Groups	925.134	6	154.189		
Total	1080.341	8			

Table A5.4.1.30: One-way ANOVA table for fresh weight on d 12 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	219.192	2	109.596	0.549	0.604
Within Groups	1197.254	6	199.542		
Total					

Table A5.4.1.31: One-way ANOVA table for fresh weight on d 14 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	391.627	2	195.813	1.067	0.401
Within Groups	1101.088	6	183.515		
Total	1492.715	8			

Table A5.4.1.32: One-way ANOVA table for fresh weight on d 16 of 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	606.303	2	303.151	1.437	0.309
Within Groups	1265.635	6	210.939		
Total	1871.938	8			

Table A5.4.1.33: One-way ANOVA table for fresh weight on d 2 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	74.519	2	37.259	10.155	0.012
Within Groups	22.014	6	3.669		
Total	96.533	8			

Table A5.4.1.34: One-way ANOVA table for fresh weight on d 4 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	75.622	2	37.811	3.055	0.122
Within Groups	74.254	6	12.376		
Total	149.876	8			

Table A5.4.1.35: One-way ANOVA table for fresh weight on d 6 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	8.107	2	4.054	0.141	0.872
Within Groups	173.044	6	28.841		
Total	181.151	8			

Table A5.4.1.36: One-way ANOVA table for fresh weight on d 8 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	17.225	2	8.613	0.160	0.856
Within Groups	322.954	6	53.826		
Total	340.179	8			

Table A5.4.1.37: One-way ANOVA table for fresh weight on d 10 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	31.440	2	15.720	0.210	0.816
Within Groups	448.337	6	74.723		
Total	479.777	8			

Table A5.4.1.38: One-way ANOVA table for fresh weight on d 12 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	85.414	2	42.707	0.435	0.666
Within Groups	589.562	6	98.260		
Total	674.976	8			

Table A5.4.1.39: One-way ANOVA table for fresh weight on d 14 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	169.283	2	84.642	0.650	0.555
Within Groups	781.241	6	130.207		
Total	950.525	8			

Table A5.4.1.40: One-way ANOVA table for fresh weight on d 16 of 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	509.031	2	254.515	1.504	0.295
Within Groups	1015.048	6	169.175		
Total	1524.079	8			

Table A5.4.1.41: One-way ANOVA table for fresh weight on d 2 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	64.616	2	32.308	8.244	0.019
Within Groups	23.515	6	3.919		
Total					

Table A5.4.1.42: One-way ANOVA table for fresh weight on d 4 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	15.039	2	7.519	0.401	0.687
Within Groups	112.589	6	18.765		
Total	127.627	8			

Table A5.4.1.43: One-way ANOVA table for fresh weight on d 6 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	6.628	2	3.314	0.298	0.753
Within Groups	66.723	6	11.121		
Total	73.351	8			

Table A5.4.1.44: One-way ANOVA table for fresh weight on d 8 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	11.737	2	5.869	0.209	0.817
Within Groups	168.793	6	28.132		
Total	180.530	8			

Table A5.4.1.45: One-way ANOVA table for fresh weight on d 10 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.514	2	1.514	0.757	0.015
Within Groups	294.413	6	294.413	49.069	
Total	295.926	8	295.926		

Table A5.4.1.46: One-way ANOVA table for fresh weight on d 12 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	4.822	2	2.411	0.037	0.964
Within Groups	389.299	6	64.883		
Total	394.121	8			

Table A5.4.1.47: One-way ANOVA table for fresh weight on d 14 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	118.133	2	59.067	0.598	0.580
Within Groups	592.585	6	98.764		
Total					

Table A5.4.1.48: One-way ANOVA table for fresh weight on d 16 of 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	176.227	2	88.114	0.738	0.517
Within Groups	716.109	6	119.351		
Total	892.336	8			

A5.4.2: Solution usage during vase life

Table A5.4.2.1: One-way ANOVA table for solution usage on d 1 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.109	2	0.054	43.434	0.000
Within Groups	0.007	6	0.001		
Total	0.117	8			

Table A5.4.2.2: One-way ANOVA table for solution usage on d 3 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.091	2	0.045	43.232	0.000
Within Groups	0.006	6	0.001		
Total	0.097	8			

Table A5.4.2.3: One-way ANOVA table for solution usage on d 5 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.053	2	0.026	10.194	0.012
Within Groups	0.015	6	0.002		
Total	0.069	8			

Table A5.4.2.4: One-way ANOVA table for solution usage on d 7 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.008	2	0.004	1.362	0.325
Within Groups	0.019	6	0.003		
Total	0.003	8			

Table A5.4.2.5: One-way ANOVA table for solution usage on d 9 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.019	2	0.009	5.619	0.042
Within Groups	0.010	6	0.002		
Total	0.029	8			

Table A5.4.2.6: One-way ANOVA table for solution usage on d 11 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.046	2	0.0234	29.704	0.001
Within Groups	0.004	6	0.0007		
Total	0.051	8			

Table A5.4.2.7: One-way ANOVA table for solution usage on d 13 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0150	2	0.00750	169.750	0.000
Within Groups	0.0002	6	0.00004		
Total	0.0153	8			

Table A5.4.2.8: One-way ANOVA table for solution usage on d 15 by 'First Red' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0240	2	0.01203	216.600	0.000
Within Groups	0.0003	6	0.00005		
Total	0.0244	8			

Table A5.4.2.9: One-way ANOVA table for solution usage on d 1 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.101	2	0.050	7.757	0.022
Within Groups	0.039	6	0.006		
Total	0.141	8			

Table A5.4.2.10: One-way ANOVA table for solution usage on d 3 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.117	2	0.058	6.697	0.030
Within Groups	0.052	6	0.008		
Total	0.169	8			

Table A5.4.2.11: One-way ANOVA table for solution usage on d 5 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.090	2	0.045	5.371	0.046
Within Groups	0.050	6	0.008		
Total	0.142	8			

Table A5.4.2.12: One-way ANOVA table for solution usage on d 7 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.078	2	0.039	4.709	0.059
Within Groups	0.049	6	0.008		
Total	0.128	8			

Table A5.4.2.13: One-way ANOVA table for solution usage on d 9 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.059	2	0.029	2.908	0.131
Within Groups	0.061	6	0.010		
Total	0.120	8			

Table A5.4.2.14: One-way ANOVA table for solution usage on d 11 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.053	2	0.026	4.235	0.071
Within Groups	0.037	6	0.006		
Total	0.091				

Table A5.4.2.15: One-way ANOVA table for solution usage on d 13 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.020	2	0.010	5.777	0.040
Within Groups	0.010	6	0.001		
Total	0.030	8			

Table A5.4.2.16: One-way ANOVA table for solution usage on d 15 by 'First Red' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.036	2	0.018	8.228	0.019
Within Groups	0.013	6	0.002		
Total	0.049	8			

Table A5.4.2.17: One-way ANOVA table for solution usage on d 1 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.077	2	0.0385	41.795	0.000
Within Groups	0.005	6	0.0009		
Total	0.082	8			

Table A5.4.2.18: One-way ANOVA table for solution usage on d 3 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0035	2	0.0177	28.500	0.001
Within Groups	0.0003	6	0.0006		
Total	0.0039	8			

Table A5.4.2.19: One-way ANOVA table for solution usage on d 5 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.023	2	0.011	9.513	0.014
Within Groups	0.007	6	0.001		
Total	0.031	8			

Table A5.4.2.20: One-way ANOVA table for solution usage on d 7 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.070	2	0.035	19.096	0.003
Within Groups	0.011	6	0.001		
Total	0.082	8			

Table A5.4.2.21: One-way ANOVA table for solution usage on d 9 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.010	2	0.005	1.628	0.272
Within Groups	0.019	6	0.003		
Total	0.027	8			

Table A5.4.2.22: One-way ANOVA table for solution usage on d 11 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.011	2	0.005	4.798	0.057
Within Groups	0.007	6	0.001		
Total	0.018	8			

Table A5.4.2.23: One-way ANOVA table for solution usage on d 13 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.007	2	0.0038	12.926	0.007
Within Groups	0.001	6	0.0003		
Total	0.009	8			

Table A5.4.2.24: One-way ANOVA table for solution usage on d 15 by 'First Red' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.018	2	0.0094	27.516	0.001
Within Groups	0.002	6	0.0003		
Total	0.021	8			

Table A5.4.2.25: One-way ANOVA table for solution usage on d 1 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.016	2	0.008	0.327	0.733
Within Groups	0.147	6	0.024		
Total	0.163	8			

Table A5.4.2.26: One-way ANOVA table for solution usage on d 3 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.087	2	0.043	1.956	0.222
Within Groups	0.134	6	0.022		
Total	0.221	8			

Table A5.4.2.27: One-way ANOVA table for solution usage on d 5 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.123	2	0.061	1.144	0.380
Within Groups	0.322	6	0.053		
Total	0.445	8			

Table A5.4.2.28: One-way ANOVA table for solution usage on d 7 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.123	2	0.061	1.144	0.380
Within Groups	0.322	6	0.053		
Total	0.445	8			

Table A5.4.2.29: One-way ANOVA table for solution usage on d 9 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.510	2	0.255	4.512	0.064
Within Groups	0.339	6	0.056		
Total	0.850	8			

Table A5.4.2.30: One-way ANOVA table for solution usage on d 11 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.035	2	0.017	0.409	0.682
Within Groups	0.260	6	0.043		
Total	0.296	8			

Table A5.4.2.31: One-way ANOVA table for solution usage on d 13 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.083	2	0.041	0.929	0.445
Within Groups	0.268	6	0.044		
Total	0.351	8			

Table A5.4.2.32: One-way ANOVA table for solution usage on d 15 by 'Akito' roses stored at 1 and 5°C and for non-stored flowers without ABA (control).

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.871	2	0.436	7.778	0.022
Within Groups	0.336	6	0.056		
Total	1.208	8			

Table A5.4.2.33: One-way ANOVA table for solution usage on d 1 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.628	2	0.314	4.235	0.071
Within Groups	0.445	6	0.074		
Total	1.074	8			

Table A5.4.2.34: One-way ANOVA table for solution usage on d 3 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1.035	2	0.518	5.296	0.047
Within Groups	0.586	6	0.097		
Total	1.622	8			

Table A5.4.2.35: One-way ANOVA table for solution usage on d 5 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.461	2	0.231	4.572	0.062
Within Groups	0.303	6	0.050		
Total	0.764	8			

Table A5.4.2.36: One-way ANOVA table for solution usage on d 7 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.461	2	0.231	4.572	0.062
Within Groups	0.303	6	0.050		
Total	0.764	8			

Table A5.4.2.37: One-way ANOVA table for solution usage on d 9 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.800	2	0.400	13.863	0.006
Within Groups	0.173	6	0.028		
Total	0.973	8			

Table A5.4.2.38: One-way ANOVA table for solution usage on d 11 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.064	2	0.032	4.311	0.069
Within Groups	0.045	6	0.007		
Total	0.110	8			

Table A5.4.2.39: One-way ANOVA table for solution usage on d 13 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.240	2	0.120	14.943	0.005
Within Groups	0.048	6	0.008		
Total	0.288	8			

Table A5.4.2.40: One-way ANOVA table for solution usage on d 15 by 'Akito' roses sprayed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.170	2	0.085	1.561	0.285
Within Groups	0.327	6	0.054		
Total	0.497	8			

Table A5.4.2.41: One-way ANOVA table for solution usage on d 1 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.087	2	0.043	1.432	0.310
Within Groups	0.182	6	0.030		
Total	0.270	8			

Table A5.4.2.42: One-way ANOVA table for solution usage on d 3 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.283	2	0.142	4.403	0.067
Within Groups	0.193	6	0.032		
Total	0.476	8			

Table A5.4.2.43: One-way ANOVA table for solution usage on d 5 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.103	2	0.051	1.882	0.232
Within Groups	0.164	6	0.027		
Total	0.267	8			

Table A5.4.2.44: One-way ANOVA table for solution usage on d 7 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.103	2	0.051	1.882	0.232
Within Groups	0.164	6	0.027		
Total	0.267	8			

Table A5.4.2.45: One-way ANOVA table for solution usage on d 9 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.531	2	0.266	12.724	0.007
Within Groups	0.125	6	0.020		
Total	0.657	8			

Table A5.4.2.46: One-way ANOVA table for solution usage on d 11 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.029	2	0.014	1.895	0.230
Within Groups	0.046	6	0.007		
Total	0.075	8			

Table A5.4.2.47: One-way ANOVA table for solution usage on d 13 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.100	2	0.050	9.705	0.013
Within Groups	0.030	6	0.005		
Total	0.131	8			

Table A5.4.2.48: One-way ANOVA table for solution usage on d 15 by 'Akito' roses pulsed with ABA and then stored at 1 and 5°C and for non-stored flowers.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.285	2	0.142	4.419	0.066
Within Groups	0.193	6	0.032		
Total	0.478	8			

APPENDIX 5.5: EFFECTS OF STORAGE TEMPERATURE AND ABA TREATMENTS ON BIOCHEMICAL ASSAYS

A5.5.1: Electrolyte leakage

Table A5.5.1.1: ANOVA table for electrolyte leakage on d 0 in petals of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	87.680	2	43.840	1.049	0.371
ABA treatment (B)	11.995	2	5.997	0.143	0.867
AxB	81.318	4	20.329	0.486	0.746
Error	752.459	18	41.803		
Total	933.452	26			

Table A5.5.1.2: ANOVA table for electrolyte leakage on d 10 in petals of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	71.467	2	35.734	1.672	0.216
ABA treatment (B)	5.431	2	2.715	0.127	0.881
AxB	75.656	4	18.914	0.885	0.493
Error	384.674	18	21.371		
Total	537.229	26			

Table A5.5.1.3: ANOVA table for electrolyte leakage on d 0 in petals of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	77.565	2	38.783	3.835	0.041
ABA treatment (B)	15.019	2	7.509	0.742	0.490
AxB	44.027	4	11.007	1.088	0.392
Error	182.046	18	10.114		
Total	318.657	26			

Table A5.5.1.4: ANOVA table for electrolyte leakage on d 10 in petals of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	1624.065	2	812.032	5.468	0.014
ABA treatment (B)	311.958	2	155.979	1.050	0.370
AxB	323.719	4	80.930	0.545	0.705
Error	2673.219	18	148.512		
Total	4932.962	26			

Table A5.5.1.5: Electrolyte leakage means on d 10 in petals of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
NOC [b]	12	12.6358		
NOS	12	15.1465	15.1465	
NOP	12	15.5689	15.5689	
5P	12	22.6548	22.6548	22.6548
5S	12	23.8291	23.8291	23.8291
1S	12	28.2679	28.2679	28.2679
1P	12	30.5896	30.5896	30.5896
5C	12		37.6004	37.6004
1C	12			39.3813
Sig.		0.128	0.061	0.151

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.1.6: ANOVA table for electrolyte leakage on d 0 in leaves of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	209.291	2	104.646	1.320	0.292
ABA treatment (B)	183.453	2	91.726	1.157	0.337
AxB	195.397	4	48.849	0.616	0.656
Error	1426.453	18	79.247		
Total	2014.594	26			

Table A5.5.1.7: ANOVA table for electrolyte leakage on d 10 in leaves of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	2333.775	2	1166.887	2.024	0.161
ABA treatment (B)	1408.905	2	704.453	1.222	0.318
AxB	563.302	4	140.825	0.244	0.909
Error	10375.043	18	576.391		
Total	14681.025	26			

Table A5.5.1.8: ANOVA table for electrolyte leakage on d 0 in leaves of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	372.371	2	186.185	14.523	0.000
ABA treatment (B)	96.653	2	48.326	3.770	0.043
AxB	123.502	4	30.875	2.408	0.087
Error	230.757	18	12.820		
Total	823.283	26			

Table A5.5.1.9: Electrolyte leakage means on d 0 in leaves of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
NOS [b]	12	6.5664			
NOC	12	8.6946			
5S	12	8.7141			
NOP	12	8.9321			
1P	12	12.1074	12.1074		
5P	12	13.2602	13.2602		
5C	12		16.0065	16.0065	
1S	12		18.1783	18.1783	
1C	12			21.1968	
Sig.		0.055	0.071	0.109	

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.1.10: ANOVA table for electrolyte leakage on d 10 in leaves of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	4224.351	2	2112.175	3.886	0.040
ABA treatment (B)	4283.537	2	2141.768	3.941	0.038
AxB	2809.130	4	702.282	1.292	0.310
Error	9782.957	18	543.498		
Total	21099.974	26			

A5.5.2: MDA

Table A5.5.2.1: ANOVA table for MDA on d 0 in petals of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	80766.240	2	40383.120	15.333	0.000
ABA treatment (B)	10692.663	2	5346.331	2.030	0.160
AxB	12606.590	4	3151.648	1.197	0.346
Error	57191.224	18	3177.290		
Total	151472.788	26			

Table A5.5.2.2: MDA means on d 0 in petals of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
NOC [b]	12	75.4075		
NOS	12	81.1942		
NOP	12	105.8908	105.8908	
1C	12	121.4167	121.4167	
1S	12	169.9833	169.9833	169.9833
5S	12		195.8167	195.8167
5P	12			225.7833
1P	12			234.0500
5C	12			235.6000
Sig.		0.056	0.063	0.175

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.2.3: ANOVA table for MDA on d 10 in petals of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	17964.155	2	8982.077	2.827	0.086
ABA treatment (B)	10560.889	2	5280.444	1.662	0.218
AxB	16728.886	4	4182.222	1.316	0.302
Error	57191.224	18	3177.290		
Total	102445.154	26			

Table A5.5.2.4: ANOVA table for MDA on d 0 in petals of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	180.155	2	90.078	10.270	0.001
ABA treatment (B)	7.837	2	3.919	0.447	0.647
AxB	13.128	4	3.282	0.374	0.824
Error	157.876	18	8.771		
Total	358.998	26			

Table A5.5.2.5: ANOVA table for MDA on d 10 in petals of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	149.681	2	74.841	5.872	0.011
ABA treatment (B)	7.031	2	3.516	0.276	0.762
AxB	19.455	4	4.864	0.382	0.819
Error	229.423	18	12.746		
Total	405.590	26			

Table A5.5.2.6: ANOVA table for MDA on d 0 in leaves of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	24010.329	2	12005.165	19.387	0.000
ABA treatment (B)	153.283	2	76.641	0.124	0.884
AxB	2858.901	4	714.725	1.154	0.363
Error	11146.030	18	619.224		
Total	38168.543	26			

Table A5.5.2.7: ANOVA table for MDA on d 10 in leaves of 'First Red' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	26809.041	2	13404.521	21.605	0.000
ABA treatment (B)	578.432	2	289.216	0.466	0.635
AxB	4000.316	4	1000.079	1.612	0.215
Error	47407.295	18	2633.739		
Total	102445.154	26			

Table A5.5.2.8: MDA means on d 10 in leaves of 'First Red' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
NOS [b]	12	66.7275		
NOC	12	69.2333		
NOP	12	79.1017	79.1017	
5P	12		115.7333	115.7333
5C	12		122.1917	122.1917
5S	12			139.2417
1S	12			140.2750
1C	12			152.6750
1P	12			160.4250
Sig.		0.573	0.059	0.065

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.2.9: ANOVA table for MDA on d 0 in leaves of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	3046.515	2	1523.257	19.120	0.000
ABA treatment (B)	445.619	2	222.809	2.797	0.088
AxB	887.858	4	221.965	2.786	0.058
Error	1434.024	18	79.668		
Total	5814.016	26			

Table A5.5.2.10: MDA means on d 0 in leaves of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
NOS [b]	12	31.4650		
NOC	12	31.7750		
NOP	12	35.8050	35.8050	
5P	12	42.9867	42.9867	
5C	12	44.4850	44.4850	
5S	12	48.3858	48.3858	
1S	12		50.2717	
1C	12		51.0983	
1P	12			75.6917
Sig.		0.052	0.077	1.000

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.2.11: ANOVA table for MDA on d 10 in leaves of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	2094.031	2	1047.016	11.804	0.001
ABA treatment (B)	326.456	2	163.228	1.840	0.187
AxB	1273.437	4	318.359	3.589	0.025
Error	1596.657	18	88.703		
Total	5290.581	26			

Table A5.5.2.12: MDA means on d 10 in leaves of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
NOC [b]	12	33.8417		
NOP	12	39.5767	39.5767	
NOS	12	42.8317	42.8317	
5P	12	47.1200	47.1200	
5S	12	48.3858	48.3858	
1S	12	51.0983	51.0983	
1C	12		52.2350	
5C	12		55.8517	
1P	12			77.5517
Sig.		0.060	0.078	1.000

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

A5.5.3: ABA

Table A5.5.3.1: ANOVA table for ABA on d 0 in petals of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	594.610	2	297.305	0.579	0.571
ABA treatment (B)	2648.002	2	1324.001	2.577	0.104
AxB	3039.626	4	759.906	1.479	0.250
Error	9246.607	18	513.700		
Total	15528.845	26			

Table A5.5.3.2: ANOVA table for ABA on d 10 in petals of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	98293.449	2	49146.724	4.148	0.033
ABA treatment (B)	273416.187	2	136708.093	11.538	0.001
AxB	302767.084	4	75691.771	6.388	0.002
Error	213272.667	18	11848.481		
Total	887749.387	26			

Table A5.5.3.3: ABA means on d 10 in petals of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
5S [b]	3	33.6667		
1S	3	44.8667		
1C	3	48.7333		
NOP	3	75.8000		
NOC	3	143.8667		
NOS	3	169.9333	169.9333	
5C	3	241.0667	241.0667	
1P	3		358.8667	358.8667
5P	3			526.4000
Sig.		0.054	0.058	0.076

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.3.4: ANOVA table for ABA on d 0 in leaves of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	1641731.518	2	820865.759	23.383	0.000
ABA treatment (B)	205915.164	2	102957.582	2.933	0.079
AxB	822998.736	4	205749.684	5.861	0.003
Error	631882.966	18	35104.609		
Total	3302528.384	26			

Table A5.5.3.5: ABA means on d 0 in leaves of 'Akito' roses separated according to Duncan's multiple range test at P = 0.05. Flowers were treated with different ABA solutions and then stored at 1 and 5°C or were not stored (control) flowers. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3
5S [b]	3	44.0000		
1S	3	44.9933		
1C	3	45.3333		
NOP	3	94.3333	94.3333	
NOC	3	163.8667	163.8667	
NOS	3	168.5333	168.5333	
5C	3	341.8000	341.8000	
1P	3		427.6000	
5P	3			1079.7333
Sig.		0.103	0.064	1.000

[a] C, 1, 5 = control, 1, 5°C. [b] C, S, P = control, spray, pulse, respectively.

Table A5.5.3.6: ANOVA table for ABA on d 10 in leaves of 'Akito' roses treated with different ABA solutions and then stored wet at low temperature.

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Storage treatment (A)	718664.354	2	359332.677	1.579	0.233
ABA treatment (B)	2563124.270	2	1281562.135	0.563	0.579
AxB	11425232.593	4	2856308.148	1.255	0.324
Error	9246.607	18	513.700		
Total	15528.845	26			

A5.5.4: Histology

Table A5.5.4.1: One-way ANOVA table for the number of vascular bundles in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	7844.016	3	2614.672	23.773	0.000
Within Groups	3519.513	32	109.985		
Total	11363.529	35			

Table A5.5.4.2: One-way ANOVA table for xylem elements per vascular bundle in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	5529.444	3	1843.148	45.967	0.000
Within Groups	1283.111	32	40.097		
Total	6812.556	35			

Table A5.5.4.3: Means of vascular bundle number in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter separated according to Duncan's multiple range test at P = 0.05. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
WINTER AK	9	36.0000			
AUTUMN AK	9		43.0000		
WINTER FR	9			59.7778	
AUTUMN FR	9				66.7778
Sig.		1.000	1.000	1.000	1.000

[a] AK, FR = 'Akito' and 'First Red', respectively.

Table A5.5.4.4: Means of xylem elements per vascular bundle in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter separated according to Duncan's multiple range test at P = 0.05. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2	3	4
WINTER AK	9	41.01111			
AUTUMN AK	9		53.34444		
WINTER FR	9			68.86667	
AUTUMN FR	9				79.75556
Sig.		1.000	1.000	1.000	1.000

[a] AK, FR = 'Akito' and 'First Red', respectively.

Table A5.5.4.5: One-way ANOVA table for vascular bundle diameter in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.013	3	0.004	3.805	0.019
Within Groups	0.036	32	0.001		
Total	0.049	35			

Table A5.5.4.6: One-way ANOVA table for vascular bundle area (%) in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	8.756	3	2.919	6.238	0.002
Within Groups	14.971	32	0.468		
Total	23.727	35			

Table A5.5.4.7: Means of vascular bundle diameter in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter separated according to Duncan's multiple range test at P = 0.05. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2
WINTER AK	9	0.3171	
AUTUMN AK	9	0.3365	0.3365
WINTER FR	9		0.3539
AUTUMN FR	9		0.3680
Sig.		0.232	0.070

[a] AK, FR = 'Akito' and 'First Red', respectively.

Table A5.5.4.8: Means of vascular bundle area (%) in transverse sections of 'First Red' and 'Akito' peduncles in autumn and winter separated according to Duncan's multiple range test at P = 0.05. Numbers on the same column are not significantly different at P = 0.05.

Treatments [a]	N	1	2
WINTER FR	9	4.3068	
AUTUMN FR	9	4.4731	
WINTER AK	9		5.1879
AUTUMN AK	9		5.5002
Sig.		0.610	0.340

[a] AK, FR = 'Akito' and 'First Red', respectively.

APPENDIX 6

EFFECT OF ABA AND ABA ANALOGUE TREATMENTS, BEFORE AND AFTER STORAGE AT 1°C, ON VASE LIFE OF CUT 'AKITO' ROSES

APPENDIX 6.1: ABA AND ABA ANALOGUE EFFECTS ON VASE LIFE

A6.1.1: Flower life

Table A6.1.1.1: ANOVA table for flower life of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	14.222	1	14.222	10.667	0.007
Vase solutions (B)	21.778	2	10.889	8.167	0.006
AxB	3.111	2	1.556	1.167	0.344
Error	16.000	12	1.333		
Total	1144.000	18			

Table A6.1.1.2: ANOVA table for foliage life of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	14.222	1	14.222	10.667	0.007
Vase solutions (B)	16.444	2	8.222	6.167	0.014
AxB	0.444	2	0.222	0.167	0.848
Error	16.000	12	1.333		
Total	1264.000	18			

Table A6.1.1.3: Means of foliage life separated according to Duncan's multiple range test at P = 0.05. Flowers were pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Treatments [a]	N	1	2	3
cc [b]	6	6.0000		
cpbi	6	7.3333	7.3333	
pc	6	8.0000	8.0000	8.0000
caba	6		8.6667	8.6667
ppbi	6		9.3333	9.3333
paba	6			10.0000
Sig.		0.066	0.072	0.072

[a] c, p = control, pulse. [b] c, aba, pbi = control, ABA, PBI-365 , respectively.

APPENDIX 6.2: ABA AND ABA ANALOGUE EFFECTS ON CHANGES IN F_v/F_m, COROLLA DIAMETER, FRESH WEIGHT AND SOLUTION USAGE DURING VASE LIFE

A6.2.1: F_v/F_m

Table A6.2.1.1: One-way ANOVA table for F_v/F_m on d 4 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00057	2	0.00028	0.405	0.684
Within Groups	0.00428	6	0.00071		
Total	0.00486	8			

Table A6.2.1.2: One-way ANOVA table for F_v/F_m on d 8 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00038	2	0.00019	0.301	0.751
Within Groups	0.00386	6	0.00064		
Total	0.00425	8			

Table A6.2.1.3: One-way ANOVA table for F_v/F_m on d 12 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00082	2	0.00041	0.939	0.442
Within Groups	0.00264	6	0.00044		
Total	0.00347	8			

Table A6.2.1.4: One-way ANOVA table for F_v/F_m on d 16 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00040	2	0.00020	0.634	0.562
Within Groups	0.00193	6	0.00032		
Total	0.00234	8			

Table A6.2.1.5: One-way ANOVA table for F_v/F_m on d 4 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00003	2	0.00001	0.081	0.923
Within Groups	0.00127	6	0.00021		
Total	0.00131	8			

Table A6.2.1.6: One-way ANOVA table for F_v/F_m on d 8 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00010	2	0.00005	1.032	0.412
Within Groups	0.00029	6	0.00005		
Total	0.00039	8			

Table A6.2.1.7: One-way ANOVA table for F_v/F_m on d 12 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00053	2	0.00026	4.377	0.067
Within Groups	0.00036	6	0.00006		
Total	0.00090	8			

Table A6.2.1.8: One-way ANOVA table for F_v/F_m on d 16 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.00116	2	0.00058	6.200	0.035
Within Groups	0.00056	6	0.00009		
Total	0.00172	8			

A6.2.2: Corolla diameter

Table A6.2.2.1: One-way ANOVA table for corolla diameter on d 0 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	34.667	2	17.333	0.258	0.781
Within Groups	403.333	6	67.222		
Total	438.000	8			

Table A6.2.2.2: One-way ANOVA table for corolla diameter on d 4 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1420.667	2	710.333	9.835	0.013
Within Groups	433.333	6	72.222		
Total	1854.000	8			

Table A6.2.2.3: One-way ANOVA table for corolla diameter on d 8 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	204.667	2	102.333	0.224	0.805
Within Groups	2735.333	6	455.889		
Total	2940.000	8			

Table A6.2.2.4: One-way ANOVA table for corolla diameter on d 12 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	261.056	2	130.528	0.350	0.718
Within Groups	2237.333	6	372.889		
Total	2498.389	8			

Table A6.2.2.5: One-way ANOVA table for corolla diameter on d 16 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	504.667	2	252.333	0.819	0.485
Within Groups	1849.333	6	308.222		
Total	2354.000	8			

Table A6.2.2.6: One-way ANOVA table for corolla diameter on d 0 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	233.556	2	116.778	0.813	0.487
Within Groups	861.333	6	143.556		
Total	1094.889	8			

Table A6.2.2.7: One-way ANOVA table for corolla diameter on d 4 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	356.222	2	178.111	0.534	0.612
Within Groups	2002.000	6	333.667		
Total	2358.222	8			

Table A6.2.2.8: One-way ANOVA table for corolla diameter on d 8 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	489.556	2	244.778	0.615	0.572
Within Groups	2389.333	6	398.222		
Total	2878.889	8			

Table A6.2.2.9: One-way ANOVA table for corolla diameter on d 12 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	505.556	2	252.778	0.888	0.459
Within Groups	1707.833	6	284.639		
Total	2213.389	8			

Table A6.2.2.10: One-way ANOVA table for corolla diameter on d 16 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	469.556	2	234.778	0.638	0.561
Within Groups	2208.667	6	368.111		
Total	2678.222	8			

A6.2.3: Fresh weight

Table A6.2.3.1: One-way ANOVA table for fresh weight on d 2 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	12.907	2	6.453	3.564	0.095
Within Groups	10.863	6	1.811		
Total	23.770	8			

Table A6.2.3.2: One-way ANOVA table for fresh weight on d 4 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	57.076	2	28.538	1.556	0.285
Within Groups	110.040	6	18.340		
Total	167.116	8			

Table A6.2.3.3: One-way ANOVA table for fresh weight on d 6 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	9.373	2	4.686	0.072	0.932
Within Groups	392.576	6	65.429		
Total	401.949	8			

Table A6.2.3.4: One-way ANOVA table for fresh weight on d 8 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	219.037	2	109.519	0.621	0.569
Within Groups	1058.591	6	176.432		
Total	1277.628	8			

Table A6.2.3.5: One-way ANOVA table for fresh weight on d 10 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	451.213	2	225.606	1.006	0.420
Within Groups	1345.636	6	224.273		
Total	1796.849	8			

Table A6.2.3.6: One-way ANOVA table for fresh weight on d 12 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1025.313	2	512.656	2.602	0.154
Within Groups	1182.292	6	197.049		
Total	2207.605	8			

Table A6.2.3.7: One-way ANOVA table for fresh weight on d 14 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1848.779	2	924.390	4.635	0.061
Within Groups	1196.658	6	199.443		
Total	3045.438	8			

Table A6.2.3.8: One-way ANOVA table for fresh weight on d 16 of 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	1865.476	2	932.738	4.822	0.056
Within Groups	1160.522	6	193.420		
Total	3025.997	8			

Table A6.2.3.9: One-way ANOVA table for fresh weight on d 2 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	21.661	2	10.831	3.976	0.080
Within Groups	16.345	6	2.724		
Total	38.006	8			

Table A6.2.3.10: One-way ANOVA table for fresh weight on d 4 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	353.587	2	176.794	2.830	0.136
Within Groups	374.882	6	62.480		
Total	728.469	8			

Table A6.2.3.11: One-way ANOVA table for fresh weight on d 6 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2359.030	2	1179.515	4.455	0.065
Within Groups	1588.649	6	264.775		
Total	3947.679	8			

Table A6.2.3.12: One-way ANOVA table for fresh weight on d 8 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2970.447	2	1485.224	6.915	0.028
Within Groups	1288.749	6	214.791		
Total	4259.196	8			

Table A6.2.3.13: One-way ANOVA table for fresh weight on d 10 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3300.332	2	1650.166	10.766	0.010
Within Groups	919.648	6	153.275		
Total	4219.980	8			

Table A6.2.3.14: One-way ANOVA table for fresh weight on d 12 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	3101.138	2	1550.569	10.985	0.010
Within Groups	846.955	6	141.159		
Total	3948.093	8			

Table A6.2.3.15: One-way ANOVA table for fresh weight on d 14 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2954.118	2	1477.059	9.097	0.015
Within Groups	974.237	6	162.373		
Total	3928.355	8			

Table A6.2.3.16: One-way ANOVA table for fresh weight on d 16 of 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	2902.397	2	1451.199	9.070	0.015
Within Groups	959.974	6	159.996		
Total	3862.371	8			

A6.2.4: Solution usage

Table A6.2.4.1: One-way ANOVA table for solution usage on d 1 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.032	2	0.016	0.600	0.579
Within Groups	0.164	6	0.027		
Total	0.197	8			

Table A6.2.4.2: One-way ANOVA table for solution usage on d 3 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.049	2	0.024	0.362	0.711
Within Groups	0.409	6	0.068		
Total	0.459	8			

Table A6.2.4.3: One-way ANOVA table for solution usage on d 5 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.084	2	0.042	1.268	0.347
Within Groups	0.200	6	0.033		
Total	0.284	8			

Table A6.2.4.4: One-way ANOVA table for solution usage on d 7 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.062	2	0.031	1.481	0.300
Within Groups	0.127	6	0.021		
Total	0.190	8			

Table A6.2.4.5: One-way ANOVA table for solution usage on d 9 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.125	2	0.062	1.213	0.361
Within Groups	0.310	6	0.051		
Total	0.435	8			

Table A6.2.4.6: One-way ANOVA table for solution usage on d 11 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.205	2	0.103	2.343	0.177
Within Groups	0.263	6	0.043		
Total	0.468	8			

Table A6.2.4.7: One-way ANOVA table for solution usage on d 13 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.213	2	0.106	2.570	0.156
Within Groups	0.248	6	0.041		
Total	0.461	8			

Table A6.2.4.8: One-way ANOVA table for solution usage on d 15 by 'Akito' roses stored at 1°C for 10 days (without pulse treatment). After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.095	2	0.047	2.728	0.144
Within Groups	0.105	6	0.017		
Total	0.200	8			

Table A6.2.4.9: One-way ANOVA table for solution usage on d 1 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.013	2	0.006	0.222	0.807
Within Groups	0.177	6	0.029		
Total	0.190	8			

Table A6.2.4.10: One-way ANOVA table for solution usage on d 3 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.157	2	0.078	7.157	0.026
Within Groups	0.065	6	0.010		
Total	0.222	8			

Table A6.2.4.11: One-way ANOVA table for solution usage on d 5 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.192	2	0.096	11.810	0.008
Within Groups	0.048	6	0.008		
Total	0.241	8			

Table A6.2.4.12: One-way ANOVA table for solution usage on d 7 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.397	2	0.198	13.479	0.006
Within Groups	0.088	6	0.014		
Total	0.485	8			

Table A6.2.4.13: One-way ANOVA table for solution usage on d 9 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.477	2	0.238	11.612	0.009
Within Groups	0.123	6	0.020		
Total					

Table A6.2.4.14: One-way ANOVA table for solution usage on d 11 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.290	2	0.145	4.062	0.077
Within Groups	0.214	6	0.003		
Total	0.504	8			

Table A6.2.4.15: One-way ANOVA table for solution usage on d 13 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.090	2	0.045	4.638	0.061
Within Groups	0.058	6	0.009		
Total	0.149	8			

Table A6.2.4.16: One-way ANOVA table for solution usage on d 15 by 'Akito' roses pulsed with 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water.

	s.s.	d.f.	m.s.	F	Sig.
Between Groups	0.0012	2	0.0006	5.435	0.045
Within Groups	0.0006	6	0.0001		
Total	0.0019	8			

APPENDIX 6.3: BIOCHEMICAL ASSAYS

A6.3.1: Electrolyte leakage

Table A6.3.1.1: ANOVA table for electrolyte leakage in petals of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	1.621	1	1.621	0.035	0.855
Vase solutions (B)	254.113	2	127.057	2.746	0.104
AxB	10.535	2	5.267	0.114	0.893
Error	555.174	12	46.265		
Total	821.443	17			

Table A6.3.1.2: ANOVA table for electrolyte leakage in leaves of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	541.791	1	541.791	1.795	0.205
Vase solutions (B)	4083.480	2	2041.740	6.764	0.011
AxB	92.577	2	46.289	0.153	0.859
Error	3622.162	12	301.847		
Total	24197.256	18			

Table A6.3.1.3: Means of electrolyte leakage in leaves separated according to Duncan's multiple range test at P = 0.05. Flowers were pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Treatments [a]	N	1	2	3
paba [b]	6	11.9303		
ppbi	6	13.8202	13.8202	
caba	6	19.8582	19.8582	
cpbi	6	31.2046	31.2046	31.2046
pc	6		46.8333	46.8333
cc	6			54.4389
Sig.		0.232	0.051	0.145

[a] c, p = control, pulse. [b] c, aba, pbi = control, ABA, PBI-365 , respectively.

A6.3.2: MDA

Table A6.3.2.1: ANOVA table for MDA in petals of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	4.646	1	4.646	1.251	0.285
Vase solutions (B)	61.800	2	30.900	8.322	0.005
AxB	7.979	2	3.989	1.074	0.372
Error	44.558	12	3.713		
Total	118.984	17			

Table A6.3.2.2: Means of MDA in petals separated according to Duncan's multiple range test at P = 0.05. Flowers were pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Treatments [a]	N	1	2	3
paba [b]	6	9.5067		
caba	6	10.3333	10.3333	
cpbi	6	10.6950	10.6950	
ppbi	6	12.9683	12.9683	12.9683
cc	6		13.6400	13.6400
pc	6			15.2417
Sig.		0.063	0.075	0.194

[a] c, p = control, pulse. [b] c, aba, pbi = control, ABA, PBI-365 , respectively.

Table A6.3.2.3: ANOVA table for MDA in leaves of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	1777.471	1	1777.471	17.176	0.001
Vase solutions (B)	888.020	2	444.010	4.290	0.039
AxB	480.439	2	240.219	2.321	0.141
Error	1241.852	12	103.488		
Total	4387.782	17			

Table A6.3.2.4: Means of MDA in leaves separated according to Duncan's multiple range test at P = 0.05. Flowers were pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Treatments [a]	N	1	2	3
ppbi [b]	6	12.1933		
paba	6		34.2033	
pc	6		36.6317	36.6317
caba	6		42.1083	42.1083
cpbi	6		45.3117	45.3117
cc	6			55.2317
Sig.		1.000	0.239	0.059

[a] c, p = control, pulse. [b] c, aba, pbi = control, ABA, PBI-365 , respectively.

A6.3.3: ABA

Table A6.3.3.1: ANOVA table for ABA in petals of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	221521.788	1	221521.788	178.777	0.000
Vase solutions (B)	91428.311	2	45714.156	36.893	0.000
AxB	64697.153	2	32348.576	26.107	0.000
Error	14869.128	12	1239.094		
Total	392516.380	17			

Table A6.3.3.2: Means of ABA in petals separated according to Duncan's multiple range test at P = 0.05. Flowers were pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Treatments [a]	N	1	2	3
cc [b]	3	89.4543		
cpbi	3	97.3964		
caba	3	117.2270		
pc	3		225.2767	
ppbi	3		235.7530	
paba	3			508.6633
Sig.		0.376	0.722	1.000

[a] c, p = control, pulse. [b] c, aba, pbi = control, ABA, PBI-365 , respectively.

Table A6.3.3.3: ANOVA table for ABA in leaves of roses pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Source of variation	s.s.	d.f.	m.s.	F	Sig.
Treatment before storage (A)	3363131.748	1	3363131.748	96.379	0.000
Vase solutions (B)	1937089.512	2	968544.756	27.756	0.000
AxB	405489.950	2	202744.975	5.810	0.017
Error	418738.502	12	34894.875		
Total	6124449.712	17			

Table A6.3.3.4: Means of ABA in leaves separated according to Duncan's multiple range test at P = 0.05. Flowers were pulsed with or without 10^{-1} M ABA and then stored at 1°C for 10 days. After storage, flowers were put into vases containing 10^{-5} M ABA, PBI-365 or distilled water (control).

Treatments [a]	N	1	2	3	4
cc [b]	3	703.9527			
cpbi	3	729.7056			
caba	3		1097.9009		
pc	3		1263.1526	1263.1526	
ppbi	3			1491.4040	
paba	3				2370.5067
Sig.		0.869	0.300	0.160	1.000

[a] c, p = control, pulse. [b] c, aba, pbi = control, ABA, PBI-365 , respectively.

Table A6.3.3.5: Parameters estimated for the linear model ($y = y_0 +/- ax$) used to describe the effects of electrolyte leakage and ABA content on flower and foliage lives of 'Akito' roses.

Temperature (°C)	Estimated parameters		Coefficient (R^2)
	y_0	a	
a. Flower life			
Electrolyte leakage	23.33	-2.049	0.61
ABA content	-387.6	77.13	0.61
b. Foliage life			
Electrolyte leakage	119.9	-10.97	0.79
ABA content	-1789.0	372.8	0.75